

Review Article

PROTAC technology for prostate cancer treatment

Zhen Wang^{a,1}, Dingpeng Zhang^{a,1}, Hiroyuki Inuzuka^a and Wenyi Wei^{a,*}

^aDepartment of Pathology, Beth Israel Deaconess Medical Center, Harvard Medical School, Boston, MA 02215, USA

¹These authors contribute equally.

*Correspondence: wwei2@bidmc.harvard.edu (W. Wei)

Received: 6 November 2024; Revised: 18 December 2024; Accepted: 11 January 2025

Published online: 30 January 2025

DOI 10.15212/AMM-2024-0075

ABSTRACT

Prostate cancer (PrCa) is the most prevalent urogenital cancer affecting men. PrCa is marked by uncontrolled cellular growth that leads to abnormal enlargement of the prostate gland. The metastatic spread of PrCa is the primary cause of mortality, causing cancer cell dissemination to distant sites, such as bones, the pelvis, and various visceral organs. Key contributors to PrCa progression include genetic mutations, elevated androgen receptor expression, gene amplification, and the rise of androgen receptor splice variants. Although androgen deprivation therapy remains the mainstay for early-stage PrCa treatment, efficacy is temporary because many cases advance to castration-resistant PrCa (CRPC), presenting a significant therapeutic hurdle. This review explores key biomarkers for PrCa and the latest therapeutic strategies for CRPC with a particular focus on the innovative proteolysis-targeting chimera (PROTAC) technology. This approach offers a novel means of degrading target proteins and we discuss how PROTAC holds potential as effective strategies to combat resistance mechanisms in CRPC.

Keywords: Chemical biology, Cancer biology, PROTAC, Prostate cancer

1. INTRODUCTION

Prostate cancer (PrCa) is characterized by uncontrolled cellular proliferation that results in abnormal growth of the prostate gland [1]. PrCa is the most common cancer affecting the male urinary and reproductive systems [1]. Metastatic PrCa cell spread to distant sites, including bones, pelvis, lumbar vertebrae, bladder, rectum, and even the brain, are the primary driver of PrCa-related mortality [2]. Notably, nearly 48% of cancer cases in men are attributed to malignancies (prostate, lung, bronchus, and colorectal cancers) with PrCa presenting approximately 27% of all diagnoses. According to the NIH, 299,010 new PrCa cases (14.9% of all new cancer cases) and 35,250 deaths occurred in the United States in 2024 [3].

Several factors contribute to PrCa progression, including genetic alterations, elevated androgen receptor (AR) expression, AR gene amplification, upregulation of cytochrome P450 17A1 (CYP17A1), and the development of AR variants [4]. Among these factors, AR splice variants (AR-Vs), particularly AR-V7 and AR-V9, lack the ligand-binding domain (LBD), rendering AR-Vs resistant to therapies targeting this domain, such as enzalutamide

and abiraterone. These AR-Vs remain active in promoting AR signaling and contribute to tumor growth and survival, even in the presence of androgen deprivation or AR-targeted therapies [5, 6]. Notably, the standard treatment for localized PrCa involves androgen deprivation therapy (ADT), which is achieved through surgical and medical castration. Although ADT can induce remission for approximately 2–3 years, PrCa often progresses to castration-resistant PrCa (CRPC), a stage that presents challenges due to limited prognosis and significant therapeutic hurdles [7].

This review provides an overview of the key PrCa biomarkers and the latest therapeutic strategies for CRPC. We place particular emphasis on the emerging proteolysis-targeting chimera (PROTAC) approach, which has garnered considerable attention for the potential to target previously “undruggable” proteins, thereby offering a promising avenue for advancing CRPC treatment [8]. PROTACs function as bispecific small molecules that facilitate proximity between target proteins of interest (POIs) and E3 ligases, thereby promoting the catalytic ubiquitination and subsequent target protein degradation. This review will delve into the various PROTACs that have been developed for PrCa,

highlighting the mechanisms of actions and potential clinical applications. Ultimately, we hope to shed light on how these innovative strategies offer promising therapeutic avenues for CRPC by targeting diverse resistance mechanism.

2. PRCA BIOMARKERS

2.1 AR

Without its ligand, AR is mainly located in the cytosol and forms complexes with heat shock proteins (HSPs) [9-11], cytoskeletal elements [12], and other chaperone proteins [10, 13]. This interaction with HSP not only stabilizes the receptor, but also induces conformational changes that prepare the AR for optimal binding when a ligand becomes available [14, 15] (Figure 1).

When AR binds to androgens like testosterone or DHT, the AR molecule undergoes a significant structural change [16]. This change prompts the AR molecule

to dissociate from HSPs, allowing AR to interact with ARA70, Filamin-A, and importin- α co-regulators. These interactions facilitate entry of AR into the nucleus, where AR predominantly functions as a homodimer. However, AR is also capable of forming heterodimers with estrogen receptor (ER) isoforms or orphan nuclear receptor testicular receptor 4 (TR4), leading to variations in transcriptional activity [10, 13, 17-22].

AR signalling is further regulated by upstream receptor tyrosine kinases (RTKs). For example, HER-2/neu and G-protein coupled receptor (GPCR) pathways can stimulate AR, even in the absence of androgen binding [23-26]. AR will undergo phosphorylation with or without ligand binding at different serine sites, subsequently affecting protein stability and transcriptional activation activity [27]. It has been reported that androgen binding specifically stimulates phosphorylation of AR at Ser64/80/93 to protect AR from degradation [28]. The phosphorylation of other residues, such as

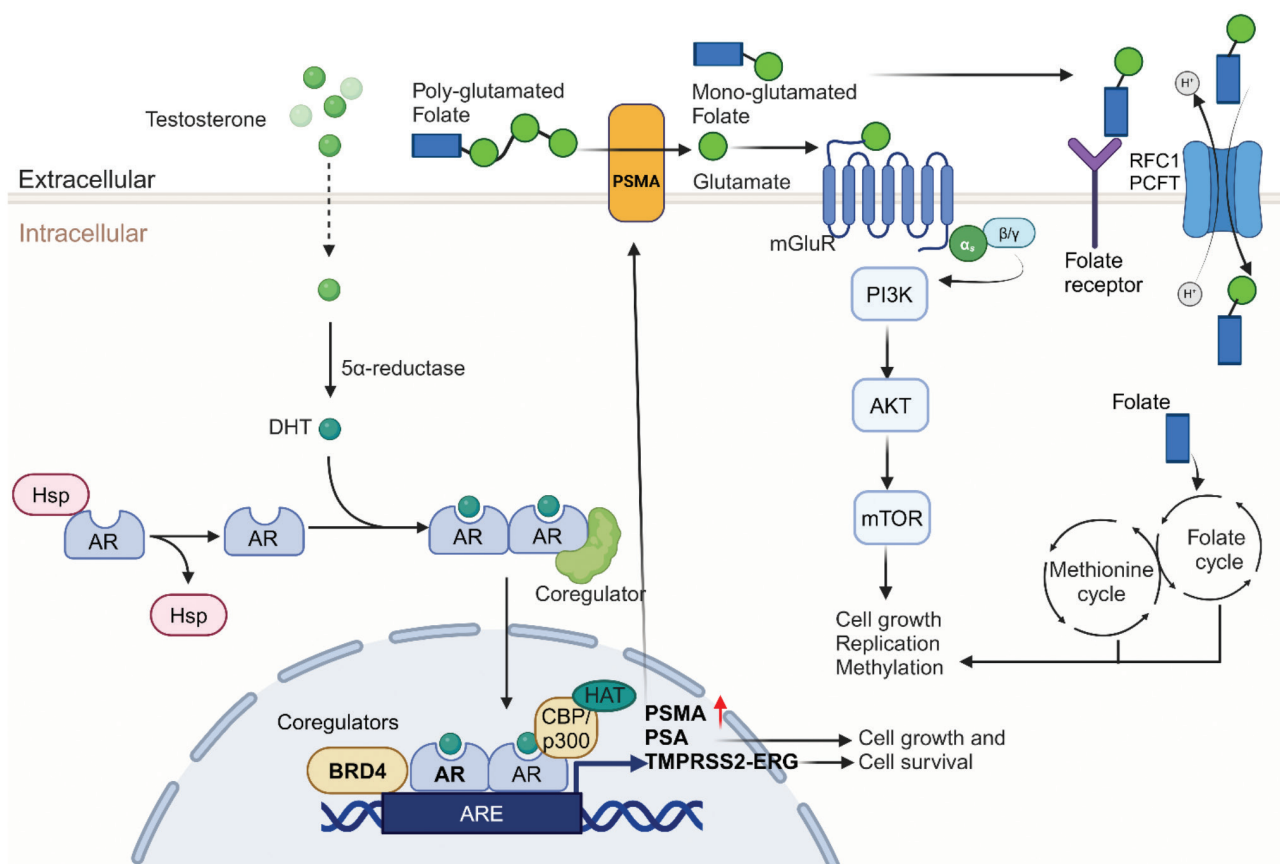


Figure 1 | The key biomarkers and the related signaling pathways in prostate cancer (PrCa).

Androgen stimulates androgen receptor (AR) signalling, resulting in the induction and secretion of prostate-specific antigen (PSMA), while illustrating the enzymatic function of prostate-specific membrane antigen PSMA and its role in folate metabolism and mGluR signaling pathways. AR, androgen receptor; DHT, dihydrotestosterone; HSP, heat shock protein; ARE, androgen response elements; BRD4, bromodomain-containing protein 4; PSMA, prostate-specific membrane antigen; PSA, prostate-specific antigen; TMPRSS2-ERG, transmembrane protease serine 2:v-ets erythroblastosis virus E26 oncogene homolog; mGluR, metabotropic glutamate receptor; PI3K, phosphoinositide 3-kinases; AKT, protein kinase B; mTOR, mammalian target of rapamycin; PCFT, proton-coupled folate transporter; RFC1, reduced folate carrier

Review Article

Ser213/506/650, via mitogen-activated protein kinases (MAPKs) is pivotal in controlling AR transcriptional activation activity. This process heightens AR sensitivity to minimal levels of androgens, estrogens, and anti-androgens by recruiting essential nuclear co-activators [29].

Upon AR nuclear translocation, the AR recognizes androgen response elements (AREs) in DNA [30]. AR recruitment of histone acetyltransferase (HAT), along with various co-regulators and core transcriptional elements, initiates target gene transcription, like PSA [18, 31, 32]. When the ligand binds to AR, the ligand will affect protein stability and transcriptional output of AR-DNA complexes. For example, when bound to antagonists, AR continues to move to the nucleus but exhibits reduced retention at AREs, which leads to diminished transcriptional activity [33, 34]. Following dissociation of the ligand, the nuclear export signal promotes the return of AR to the cytosol for preparation of another cycle of ligand binding or proteasomal degradation, thereby maintaining overall AR activity [18, 35-38].

2.2 Prostate-specific membrane antigen (PSMA)

The *FOLH1* gene is known to encode PSMA [39, 40]. PSMA is referred to as N-acetyl-L-aspartyl-L-glutamate peptidase I (NAALADase I) in the fields of neurology and metabolism [41, 42]. Despite these variations in nomenclature, PSMA acts as a glycoprotein on the cell membrane, catalyzing the hydrolysis of folate and carboxypeptides [43, 44] (Figure 1). PSMA has a critical role in processing dietary folates and is primarily distributed in the prostate, kidneys, and duodenum. Interestingly, PSMA is also present in low levels within brain tissue, where PSMA stimulates activation of metabotropic glutamate receptors (mGluRs) [42].

PSMA is comprised of an intracellular domain, a transmembrane segment, and a large extracellular domain [43, 45]. By cleaving glutamated folate, PSMA is responsible for releasing glutamate, the predominant dietary form of folate [46] (Figure 1). Following the enzymatic action of PSMA, the generated mono-glutamated folate is then absorbed by transporters, such as the folate receptor (FR), reduced folate carrier (RFC)1, and proton-coupled folate transporter (PCFT) [47]. Folate uptake is especially advantageous for rapidly dividing cells because folate supports many cellular processes, including one-carbon metabolism, nucleotide synthesis, and epigenetic methylation, all of which intersect with the methionine cycle [48, 49] (Figure 1). Additionally, under conditions of limited folate availability, hydrolysis of folate by PSMA might confer a growth advantage [50].

AR and neuroendocrine PrCa (NEPC) modulate the one-carbon metabolic pathway, which is closely intertwined with folate-mediated transfer [51-53]. Within the mammalian nervous system, N-acetyl-aspartyl-glutamate (NAAG) is among the most abundant neuropeptides and is particularly present at neuronal synapses [54, 55]. NAAG triggers activation of mGluR3

and is subsequently broken down by PSMA. By releasing glutamate, PSMA promotes phosphatidylinositol 3-kinase-AKT-mammalian target of rapamycin (PI3K-AKT-mTOR) signaling [56]. First, glutamate release initiates activation of mGluR I, which leads to phosphorylation of p110 β and activation of PI3K. Activated PI3K, in turn, stimulates the AKT-mTOR pathway, driving protein synthesis and cellular proliferation to facilitate tumorigenesis [57] (Figure 1). Notably, preclinical studies have shown that the PSMA inhibitor, 2-PMPA, effectively suppresses PI3K signaling, resulting in tumor regression [56]. Moreover, elevated levels of mGluRs are recognized as a possible adaptive strategy to maintain glutamatergic activity in NEPC [58].

PSMA-regulated folate metabolism impacts a variety of cellular processes, including tumor cell proliferation, DNA repair, drug resistance, and resource prioritization in tumor microenvironments [59-61]. As a result, PSMA contributes to tumor growth and proliferation by increasing folate availability, which drives anabolic metabolic pathways essential for cellular development and division [59]. Additionally, PSMA-regulated folate metabolism is closely linked to DNA repair mechanisms in cancer [60]. Metabolic intermediates, such as methylene tetrahydrofolate produced through one-carbon metabolism, are essential for thymidine synthesis and DNA repair [60]. Disruption of this pathway may result in genomic instability and address drug resistance. Moreover, PSMA regulates resource prioritization in the tumor microenvironment. By enhancing folate utilization, PSMA provides cancer cells with a competitive advantage to sustain high proliferation rates within nutrient-deprived tumor microenvironments [61]. This metabolic adaptation is believed to contribute to the aggressive phenotype of PSMA-positive tumors. Notably, PSMA upregulation corresponds to the increased metabolic demands of tumor cells, which allows tumor cells to outcompete surrounding normal cells for access to folate and related resources [61].

2.3 Bromodomain-containing protein 4 (BRD4)

Epigenetic readers, including BRD4, have been shown to be key molecules associated with transcription factors (TFs), like AR, and contribute to the development of aggressive cancers, including PrCa [62-66]. The detailed mechanisms by which bromodomain inhibitors exert an effect remain largely undefined [64]. Recent research has shown that changes in DNA accessibility effectively distinguish PrCa from early-stage malignancies and non-cancerous tissues. The increased genomic accessibility observed in advanced tumors appears to be mediated, at least in part, by indirect pathways involving AR interaction with BRDs (BRD4, BRD2, and ATAD2), which have been identified as key facilitators contributing to enhanced accessibility. Notably, these proteins are overexpressed in CRPC and serve as prognostic tissue markers, underscoring their potential as targets in aggressive PrCa [67].

2.4 Prostate-specific antigen (PSA)

PSA, a serine protease, is secreted by prostate epithelial cells [68]. Although naturally occurring in normal prostatic secretions, PSA is frequently increased in PrCa patients [69, 70]. For over 2 decades, the serum PSA level has been served as a crucial biomarker in the detection of PrCa [71]. There has been a significant improvement in PrCa management since the 1980s with the introduction of PSA screening, which has contributed to better survival rates. Despite these advances, PSA has limitations as a biomarker, which has prompted the exploration of more accurate diagnostic tools and accelerated the development of PrCa biomarkers [71, 72].

2.5 Transmembrane protease serine 2: v-ets erythroblastosis virus E26 oncogene homolog (TMPRSS2-ERG)

A promising biomarker in PrCa diagnostics is the TMPRSS2-ERG fusion. ETS family transcription factors, such as ERG, when fused with the androgen-regulated *TMPRSS2* gene represent the most frequently observed gene fusions, accounting for approximately 50% of PrCa cases [73, 74]. A study by Laxman et al. [75] first reported the presence of gene fusion events in the patients with PrCa. This discovery has since been supported by additional studies that indicate *TMPRSS2-ERG* fusions could function as a urinary biomarker. Notably, these gene fusions exhibit high specificity and a strong positive predictive value. However, the sensitivity of *TMPRSS2-ERG* fusions is relatively low [76, 77].

Because most tumors consist of multiple distinct foci, the tumor heterogeneity characteristic of PrCa is a significant limitation to the use of this gene fusion as a biomarker [76]. Moreover, the prognostic value of *TMPRSS2-ERG* fusion has not been established. Although some studies have shown that this gene fusion correlates with increased cancer aggressiveness, metastasis, and mortality [78, 79], other studies have not confirmed a consistent relationship between this fusion and clinical outcomes, which warrants further in-depth investigations [80].

3. TARGETED THERAPIES FOR PRCA BIOMARKERS

The current treatment landscape for PrCa is progressive and adaptable and the strategies evolve according to the stage and status of the disease. PrCa is often detected through early screening methods in the initial diagnosis, with treatments typically including surgical removal of the prostate or radiotherapy. These initial treatments eliminate the tumor confined to the prostate. However, recurrence remains a concern because approximately 30% of PrCa patients experience a return of cancer following local therapy [81] (Figure 2A).

Disease recurrence is diagnosed based on elevated PSA levels or imaging techniques that reveal the presence of residual or metastatic disease. When facing biochemical or localized recurrence, physicians may choose to

closely monitor PrCa patients without resorting to ADT immediately to avoid the associated side effects. ADT is withheld until disease progression reaches a pre-established threshold. Once this threshold is reached, ADT is typically initiated using luteinizing hormone-releasing hormone (LHRH) antagonists or agonists to decrease androgen levels and inhibit cancer growth. This treatment regimen may be administered intermittently with treatment holidays to help manage side effects until evidence of disease progression occurs [82].

While ADT can initially achieve disease control, many PrCa patients ultimately become resistant to this therapy and progress to a more advanced form of PrCa (CRPC). This progression marks a turning point in the treatment regimen because CRPC no longer responds to traditional hormone therapies and requires alternative strategies. In cases in which PrCa has already metastasized beyond the prostate gland before the initiation of local treatments, a more aggressive approach is necessary. To this end, physicians and patients must then decide on the early use of ADT, either in conjunction with radiotherapy [83] or docetaxel [84].

As patients progress through these treatments the tumor acquires resistance to therapies targeting AR but a range of additional treatment options exist. These options include poly(ADP-ribose) polymerase (PARP) inhibitors, which take advantage of DNA repair deficiencies in cancer, and chemotherapy regimens designed to target rapidly dividing tumor cells [85]. Additionally, PSMA-targeted treatments represent a novel and promising option by selectively targeting PSMA-expressing PrCa cells [82]. Moreover, immunotherapy represents an expanding avenue for advanced PrCa by harnessing the immune system to attack cancer cells [82]. This extensive array of treatment strategies, as illustrated in Figure 2, highlights the complexity and adaptability required in managing PrCa.

3.1 BRD4 targeted inhibitors

The molecular landscape in PrCa is highly intricate, with various signaling pathways and genetic mutations contributing to disease progression and drug resistance. One promising therapeutic approach in PrCa targets BRD4, which is implicated in cancer cell survival and proliferation [86, 87]. NEO2734, a novel dual inhibitor, has been developed to target both bromodomain and extra-terminal (BET) proteins (including BRD4) and CBP/p300, a family of transcriptional co-activators with histone acetyltransferase activity. NEO2734 represents an innovative treatment strategy, especially for the most frequent mutated gene in PrCa, *Speckle-type POZ protein (SPOP)* [88]. Under normal circumstances, wild-type *SPOP* is pivotal in regulating cellular protein levels by binding to and promoting BET protein degradation. However, this regulatory function is lost when *SPOP* is mutated, leading to the excessive accumulation of BET proteins. The subsequent dysregulation of gene expression pathways drives cancer growth. This loss of function

Review Article

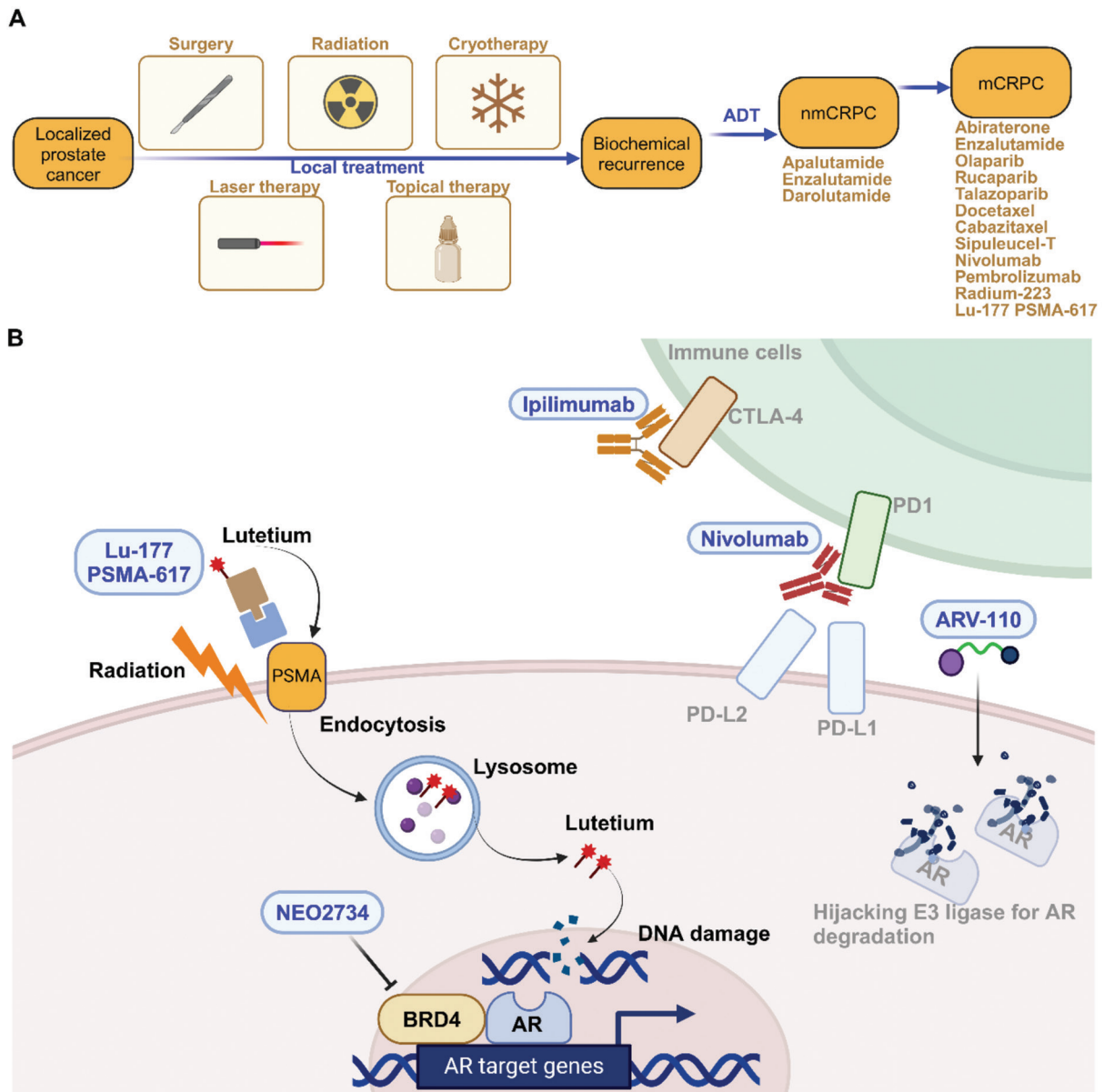


Figure 2 | Current therapy landscape for PrCa.

(A). Available treatment options for different stages of PrCa. (B). Various therapies targeting BRD4, AR, PSMA, and immune check points in PrCa. ADT, androgen deprivation therapy; nmCRPC, non-metastatic castration-resistant PrCa; mCRPC, metastatic castration-resistant PrCa; Lu, lutetium; AR, androgen receptor; BRD4, bromodomain-containing protein 4; PD-L1, programmed cell death-ligand 1; PD-L2, programmed cell death-ligand 2; CTLA-4, cytotoxic T-lymphocyte antigen 4.

often results in drug resistance against BET inhibitors (BETis) in *SPOP*-mutant PrCa patients [88].

Preclinical studies have reported that NEO2734 significantly inhibits growth of *SPOP*-mutant PrCa [88]. Furthermore, NEO2734 has shown efficacy against enzalutamide-resistant PrCa cells and patient-derived organoids, indicating the potential use as a treatment strategy for cases that have developed resistance to anti-androgen therapies [89]. Recent research has also

explored the function of AR splice variants (AR-Vs) in conferring increased castration resistance in PrCa. It has been observed that NEO2734 counteracts the mechanisms associated with anti-androgen-induced ferroptosis [90]. By inhibiting this process, NEO2734 offers a promising strategy for overcoming resistance that restricts the effectiveness of existing therapies [90]. Currently, NEO2734 is being investigated in clinical trials for CRPC patients (NCT05488548). This trial aims to evaluate the

safety, tolerability, and preliminary efficacy of NEO2734 in CRPC patients, which offers hope for a novel therapeutic option that targets multiple components of the complex molecular machinery driving PrCa progression.

3.2 PSMA targeted therapy: Lu 177-PSMA-617

Pluvicto (177Lu-PSMA-617) is another promising therapeutic option for targeting androgen receptor variant-containing cells, especially those cells that also display PSMA. This innovative treatment consists of a molecule targeting PSMA conjugated to 177Lu, a radioactive isotope. The mechanism of action enables 177Lu-PSMA-617 to selectively bind PSMA-expressing PrCa cells, facilitating the precise delivery of radiation to the tumor without damaging normal tissues. As such, 177Lu emits radiation that directly harms the DNA of cancer cells, ultimately leading to cell death [91]. This precision-targeted approach has yielded highly promising outcomes, especially in advanced-stage PrCa, as demonstrated in the NCT03511664 clinical trial [92].

Recent studies have explored molecular biomarkers in patients treated with Pluvicto. One study analysed circulating tumor cells and investigated clinical factors, such as protein levels of PSA and PSMA expression, tumor size, and the levels of AR and AR-V7 expression [5]. The findings revealed that AR and AR-V7 expression could be used as prognostic markers in metastatic CRPC patients before starting Pluvicto therapy. Interestingly, while these biomarkers correlated with tumor load, the biomarkers did not predict the patient's response to PSMA-targeted therapies. Notably, AR-V7 was associated with higher AR expression, increased tumor load, and an elevated level of PSMA protein [5].

Alternatively, Pathmanandavel et al. [93] reported that among AR-V7-positive metastatic (m)CRPC patients treated with 177Lu-PSMA-617, AR-V7 did not adversely impact overall survival. This observation suggested that 177Lu-PSMA-617 could be a viable therapy for mCRPC patients expressing AR-V7 along with established treatments, such as taxanes. However, exploring the function of protein expression and the potential value of these markers in determining treatment outcomes is important in further research.

The therapeutic potential of 177Lu-PSMA-617 continues to be investigated across multiple ongoing clinical trials. The PSMA fore-trial (NCT04689828) aims to compare the efficacy of 177Lu-PSMA-617 therapies in mCRPC patients targeting the AR. Another active trial is investigating pairing Pluvicto with pembrolizumab, an immune checkpoint inhibitor [ICI] (NCT03805594) [94, 95].

3.3 Immunotherapy: nivolumab and ipilimumab

ICIs, including nivolumab and ipilimumab, have been shown to be a potential treatment strategy for advanced PrCa, especially in AR-V7+ PrCa patients. These inhibitors target immune response regulators,

effectively enhancing the ability of immune cells to eliminate cancer cells [96]. Nivolumab targets immune cells by blocking the programmed cell death protein (PD)-1 receptor. Under normal conditions, PD-1 binds to physiologic ligands (PD-L1 and PD-L2) expressed on the cancer cell surface, resulting in suppression of the immune reaction and allowing the cancer cells to escape immune surveillance. Blocking this interaction, nivolumab reactivates the immune response, allowing the immune response to recognize and attack tumor cells more effectively. In contrast, ipilimumab targets another checkpoint molecule, cytotoxic T-lymphocyte antigen 4 (CTLA-4), a membrane receptor on immune cells that suppresses immune activity. By inhibiting CTLA-4, ipilimumab prevents the inhibitory signaling pathway, which keeps the immune cells active and enables tumor cells to target cancer cells [96].

In clinical practice nivolumab and ipilimumab are frequently used together to create a synergistic immune reaction towards cancer cells. NCT02601014, a clinical trial evaluating this combination in AR-V7+ PrCa patients, demonstrated promising efficacy. However, the response was not consistent across AR-V7+ patients [97].

The efficacy of combining nivolumab and ipilimumab, either along with or independent of enzalutamide, in AR-V7+ mCRPC patients was investigated in the same phase II trial. While combination therapy had an acceptable profile regarding safety, combination therapy had limited efficacy in this patient population, even when combined with enzalutamide [98]. Further insights were gained from another phase II trial (NCT02985957). Preliminary findings suggested that some CRPC patients experience complete responses to the combination of nivolumab and ipilimumab, with four participants achieving complete remission [99]. However, the study did not establish a consistent correlation between treatment efficacy and tumor mutation burden in blood or tissue samples [99].

Numerous clinical trials are actively exploring the combination of ICIs with other therapeutic agents. These trials involved the combination of 177Lu-PSMA (NCT05150236) and stereotactic body radiotherapy [SBRT] (NCT05655715). Another promising immunotherapy for metastatic PrCa is the DNA vaccine, MVI-118, which encodes the ligand-binding domain (LBD) of the AR and induces the immune response against AR-overexpressing cancer cells mediated by CD8+ T. MVI-118 was shown to be safe and effective in NCT02411786 with respect to activating the immune system in PrCa [100]. Two other clinical trials are currently investigating the MVI-118. One trial is exploring the use of the pTVG-HP DNA vaccine with or without MVI-118 and pembrolizumab in mCRPC patients, while the other trial is examining ADT with or without MVI-118 and/or nivolumab in PrCa patients, according to the NCT04090528 and NCT04989946 clinical trials, respectively.

Review Article

3.4 Limitations of current approaches

Despite the development of numerous targeted therapies aimed at PrCa biomarkers, these strategies often encounter substantial challenges, which highlight the pressing need for novel therapeutic innovations. PROTAC technology has emerged as a transformative approach, offering unique advantages that address key limitations associated with conventional methods [8]. Compared to the aforementioned therapeutic modalities, PROTACs overcome several notable constraints, underscoring the potential of this technology to redefine treatment paradigms for PrCa.

3.4.1 Resistance to AR inhibitors. Enzalutamide and abiraterone, the current standard AR inhibitors, are effective in prolonging survival [101–104]. However, several problems, including AR gene amplification, overexpression of splice variants with constitutive activity (e.g., AR-V7), mutations in the AR LBD, and activation of compensatory signaling pathways, contribute to resistance mechanisms and limit clinical efficacy [5, 6]. Compared to the inhibitors, PROTACs offer a unique approach by degrading AR proteins, including splice variants and mutant forms, and may potentially overcome these resistance mechanisms [8].

3.4.2 Toxicity and side effects of chemotherapy. Chemotherapy remains a standard treatment option for advanced CRPC, including docetaxel and cabazitaxel for PrCa therapy [105]. However, these agents have been reported to exhibit significant toxicities, such as neutropenia, neuropathy, and fatigue, which limit long-term use [106]. PROTACS may offer a safer therapeutic alternative with fewer off-target effects, in part due to the specificity and targeted mechanism of action.

3.4.3 Mechanistic limitations of conventional therapies. The mechanisms of action of traditional therapies primarily focus on inhibiting AR signaling [106] rather than eliminating the functional AR, which results in drug resistance. In contrast, PROTACs aim to entirely degrade the AR protein, the key driver of CRPC progression. This degradation likely disrupts downstream transcriptional activity and reduces the likelihood of reactivation.

3.4.4 Addressing unmet needs. PROTACs are designed to target proteins beyond AR that are involved in CRPC progression, thereby expanding the therapeutic scope and enabling personalized treatment strategies based on specific molecular profiles. However, achieving this goal may be challenging with traditional inhibitors.

In conclusion, the unique therapeutic value of PROTAC technology lies in an ability to address the key challenges of resistance, toxicity, and limited mechanistic action, which underscores the potential in treating CRPC.

3.5 PROTACs towards AR

Among the innovative therapeutic strategies under investigation for PrCa, PROTACs have emerged as promising strategies to selectively degrade target proteins (POI) [8]. One of the leading PROTACs in clinical development is ARV-110, which became the first PROTAC used in clinical trials by targeting AR for destruction. By leveraging the ubiquitin-proteasome system (UPS), ARV-110 specifically induces AR degradation [107].

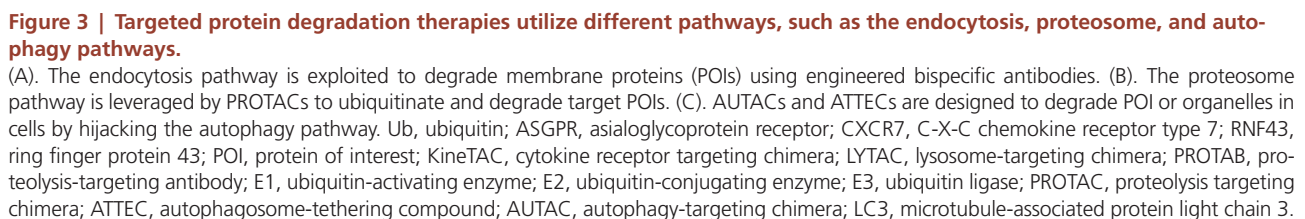
The mechanism by which ARV-110 functions involves forming a ternary complex that induces the AR proximity between POI with an E3 ligase (specifically, cereblon [CRBN]) [8]. CRBN consists of two distinct components linked by a flexible linker. One side features a non-covalently binder that binds to the AR and the other side contains a thalidomide-derived “warhead” that covalently attaches to the CRBN. This binding induces the formation of the E3 ligase-ARV-110-AR complex, consisting of the E3 ligase, ARV-110, and the AR protein. By bringing AR and CRBN into proximity, ARV-110 facilitates tagging of ubiquitin molecules to AR, ultimately leading to degradation (Figure 2B). This process results in complete degradation of the AR, rather than merely inhibiting the AR, thereby significantly reducing the AR level within cancer cells.

What makes ARV-110 particularly innovative is that, unlike traditional AR inhibitors that simply block AR activity, ARV-110 actively triggers the AR degradation. This mechanism is advantageous, especially in cases in which PrCa cells have developed resistance to conventional AR therapies. By degrading, rather than merely inhibiting the AR protein, ARV-110 offers a potential solution to overcoming resistance mechanisms arising from mutations in the AR LBD, which often reduce the treatment efficacy of standard therapies. Another essential advantage of ARV-110 is the catalytic mechanism. The free PROTAC and E3 ligase complex can be recycled and participate in the next round of degradation. This catalytic mechanism renders ARV-110 a highly efficient therapeutic agent, offering superiority over traditional inhibitors in a stoichiometric manner. By exploiting the UPS to degrade AR, ARV-110 represents a new frontier in targeted cancer therapy, potentially offering a more effective treatment method for PrCa patients [8].

4. PROXIMITY STRATEGIES

4.1 Membrane protein degradation strategy

Endocytosis has an important role in the macromolecule uptake pathway (Figure 3A). The internalized macromolecules bind to the specific receptors on cell surface and accumulate in the clathrin-coated pits. After detaching from the membrane, these pits facilitate the enclosure of receptors and corresponding ligands through forming vesicles. The vesicles will subsequently merge with early endosomes and are then directed to lysosomes or back to the cell surface for recycling [108].



disease-associated membrane protein degradation, revolutionizing the treatment landscape by rendering previously “undruggable” targets amenable to therapeutic intervention.

In addition to endocytosis, a key mechanism for regulating protein levels in eukaryotic cells is ubiquitination, followed by targeted protein degradation. Cellular ubiquitination-induced proteolysis is a pathway in which proteins undergo polyubiquitination, then degradation by the proteasome or lysosome [114-117]. This mechanism is precisely controlled and consists of a cascade of enzymatic activities, including E1, E2, and E3, to facilitate attachment of ubiquitin to the substrate protein lysine residues

Review Article

[118]. Importantly, the ubiquitination process is reversible because deubiquitinases (DUBs) cleave polyubiquitin chains from modified proteins, thereby allowing for dynamic regulation of protein stability in cells [119].

Various types of polyubiquitin chains are formed, each serving distinct biological functions, due to the presence of seven lysine residues in ubiquitin protein. For example, two polyubiquitin chains (K11 and K48) primarily tag proteins, then degrade the proteins, while K63-linked chains typically function as docking sites for protein-protein interactions, facilitating signal transduction events [116, 117]. The specificity of the ubiquitination process is controlled by E3 ligase [120]. However, a limited number of these ligases have clearly defined degron recognition capabilities among > 600 E3 ligases [121]. Degron recognition is a crucial process for removing misfolded or damaged proteins and maintaining intracellular protein balance. Targeted protein degradation (TPD) that selectively degrades pathogenic proteins is a promising therapeutic strategy.

PROTACs, novel bifunctional molecules, are engineered to exploit the ubiquitin-proteasome system for the degradation of POIs [122]. This kind of design allows PROTACs to form a ternary structure between the POI and an E3 ligase, facilitating targeted protein ubiquitination and degradation (Figure 3B). Notably, an AR targeting the PROTAC compound, ARV-110, is currently undergoing clinical trials for treating PrCa [8].

4.3 AUTACs strategy

Recent advances in autophagy-based degraders have introduced two notable examples (autophagy-targeting chimeras [AUTACs] and autophagosome-tethering compounds [ATTECs]). AUTACs represent one of the pioneering approaches in this field, as reported by Takahashi et al. [123, 124]. ATTECs developed by Li et al. [125, 126] function as molecular glues that mimic autophagy adaptors. This model involves the targeted degradation of specific proteins via autophagy, in which ATTECs facilitate the process by acting similar to natural autophagy adaptors (Figure 3C). While ATTECs and AUTACs have not been used for AR degradation, these platforms hold significant promise for future applications in CRPC therapy.

5. PROTAC STRATEGIES

5.1 Current PROTACs with different component designs

A PROTAC molecule consists of three essential parts (the E3 ligase ligand, a POI ligand, and a linker that connects these two ligands). Each of these elements is crucial in facilitating the targeted degradation. The detailed characteristics are discussed in the following sections.

5.1.1 PROTAC with various E3 ligases. Various E3 ligases, such as β -transducin repeat-containing protein (β -TrCP) [127], von Hippel-Lindau (VHL) [128], MDM2 [129],

CRBN [130], and cellular inhibitor of apoptosis protein (cIAP)1 [131] have been used in PROTAC development. The catalytic nature of PROTACs allows PROTACs to act repeatedly, making PROTACs highly effective in POI degradation [132, 133].

5.1.2 PROTAC with different linkers. Figure 4A provides a classification of the most frequent used linkers in according to the structural data in the database [134]. The current linker categories include flexible linkers, rigid linkers, triazole-based linkers, bio-orthogonal clickable linkers, and photo-switchable linkers.

A common strategy in modifying the linkers to enhance the solubility of PROTACs involves incorporating heterocycles (saturated) with a basic center. Examples, such as the piperidine or piperazine rings, belong to the category of rigid linkers frequently used in PROTAC design (Figure 4A) [135-137]. The inclusion of piperidine or piperazine not only contributes to the increased rigidity of the PROTAC, which can potentially enhance PROTAC activity but also introduces a protonable amino group that may improve solubility. Piperazine-containing linkers offer the advantage of enhancing the overall solubility of the molecule. However, data from the PROTAC-DB, an open-access database compiling PROTAC information, indicates that the pKa of an acidic or basic center within a molecule can be significantly influenced by neighbouring groups [138, 139] and affecting PROTAC properties.

5.1.3 PROTAC targets of different POIs. The ideal targets for PROTAC therapy typically possess a set of common characteristics, especially alterations from their natural state, such as overexpression, mutation, protein aggregation, and changes in isoform expression. Additionally, an optimal target should feature an accessible surface that allows E3 ligase binding. Furthermore, the presence of a flexible domain that can facilitate efficient and effective translocation into the proteasome is considered advantageous for facilitating protein degradation [139, 140].

5.1.3.1 Inflammation, immunity, and immuno-oncology

Two promising candidates, a Bruton's tyrosine kinase (BTK) degrader (NX-5948) [141], and an IRAK4 degrader (KT-474) [142], are currently being investigated to treat a range of immuno-inflammatory conditions in clinical trials. There is a highly validated therapeutic target-BTK in inflammatory diseases and oncology with several approved treatments already available [143, 144]. However, the rise in the C481S resistance mutation in BTK has diminished the efficacy of first-generation BTK inhibitors, driving significant interest in developing PROTAC-based therapies to address this challenge [145-147]. PROTACs offer a key advantage through event-driven pharmacology, enabling PROTACs to catalytically degrade target proteins rather than merely inhibit target proteins. This mechanism holds promise

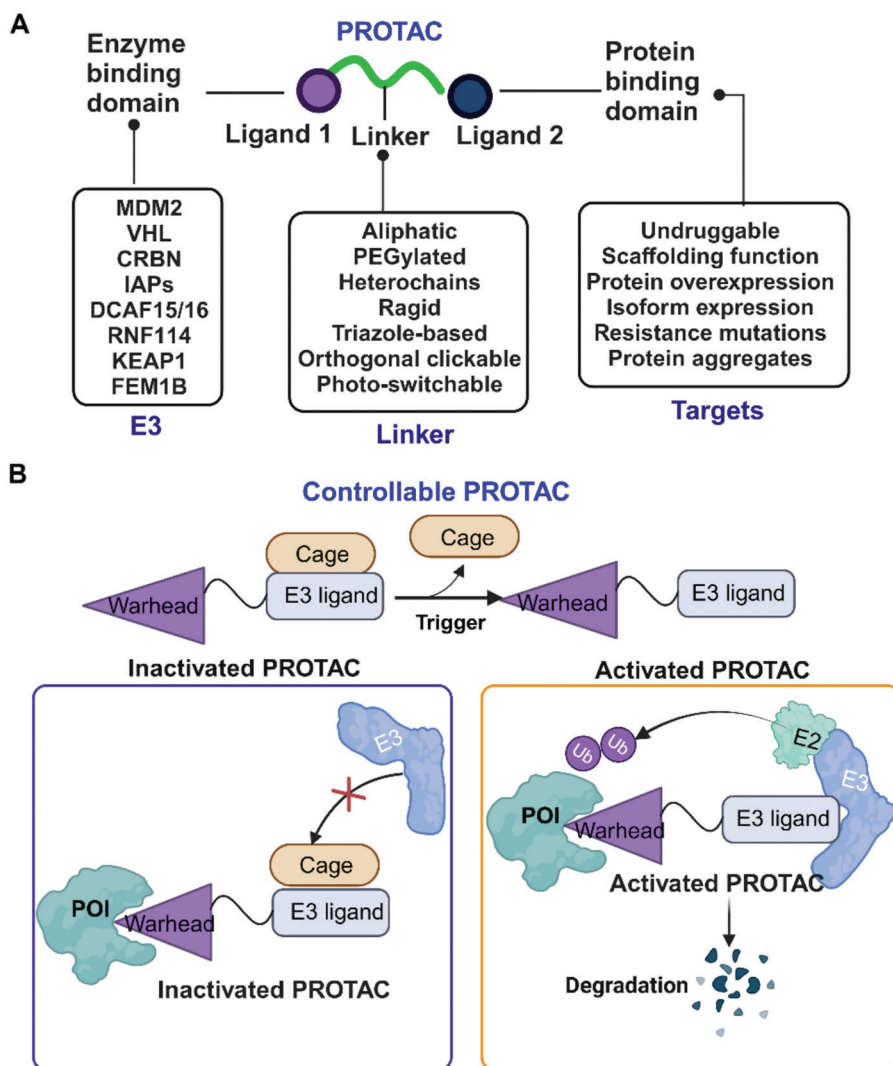


Figure 4 | PROTAC construct and the mechanism for controllable PROTAC.

(A). The PROTAC construct, which includes E3 ligase ligands, target protein ligands, and the linkers connecting the E3 ligase and target protein ligands. The reported types of E3 ligases, linkers, and target proteins in PROTACs are summarized in the indicated boxes. B. The design and working mechanism of controllable PROTACs.

for overcoming resistance mutations like C481S, which reduce the potency of conventional, occupancy-driven inhibitors [148, 149].

5.1.3.2 Neurology and neurodegeneration

One breakthrough in PROTAC technology is the ability to degrade undruggable proteins. These proteins typically cannot be targeted by inhibitors due to a lack of binding sites. The advantage of PROTAC make it possible to address these disease-causing proteins, especially in neurodegenerative diseases, such as tau [150–157], α -synuclein [158, 159], and mutant huntingtin (mHTT) [125, 160–162].

Lu et al. [154] identified Peptide 1, a KEAP1-dependent PROTAC, which was shown to degrade tau

protein, highlighting the potential in the future application. In 2020, Qu et al. [159] introduced another TAT-PBD-PTM based on peptides that specifically targets and degrades α -synuclein in Parkinson's disease [159]. Additionally, Tomoshige [162] designed hybrid small molecules (compounds 1 and 2), which effectively degrade mHTT, demonstrating promise for the treatment of Huntington's disease [162].

5.1.3.3 Transcription factor

Transcription factors and RNA-binding proteins (RBPs) have critical roles in DNA integrity, synthesis, gene regulation, and diverse RNA-mediated mechanisms [163–165]. Despite the importance of transcription factors and RBPs, many of these POIs without ligand-binding

Review Article

sites are notoriously challenging to target with traditional drugs [166, 167]. To address these “undruggable” targets, various studies have been conducted to establish innovative approaches. For example, transcription factors have been targeted using strategies, such as transcription factor-targeting chimera (TRAFTAC) [168], transcription factor PROTAC (TF-PROTAC) [169], oligonucleotide-PROTAC (O’PROTAC) [170], and G-quadruplex PROTAC (G4-PROTAC) [171]. Beyond transcription factors, telomeres have been targeted using telomere-targeting chimera (TeloTAC) [172], showcasing the potential to expand to more sophisticated methodologies. Additional advanced strategies include developing Myc-targeting PROTACs using a TNA-DNA bivalent binder [173], MeCP2 degraders that utilize methylated DNA [174], and structurally specific Z-DNA PROTACs for targeting Z-DNA binding proteins [175]. These DNA- or RNA-based PROTACs use consensus RNA or DNA sequences, which are linked to an E3 ligase ligand. This innovative approach significantly broadens the possibilities for targeting previously undruggable proteins within cells, offering new avenues for therapeutic intervention.

5.2 Current controllable PROTACs

Several controllable strategies for PROTACs have been reported, as summarized in Table 1, including stimuli-responsive approaches, such as photoactivation, folate targeting, hypoxia sensitivity, X-ray induction, and other external triggers.

5.3 Recent developments in AR degradation technology

AR degraders utilizing the PROTAC principle have facilitated significant progress with a growing number of

unique AR degraders being synthesized and tested for efficacy against PrCa against PrCa (Table 2).

CRBN-based PROTACs, especially ARV-110, are among the most widely studied PROTACs for AR degradation [8]. One notable study modified the FDA-approved AR inhibitor, enzalutamide, using various scaffolds in combination with different linkers. The most promising compound from this subseries, PROTAC 35 (compound 1), exhibited potent degradation capabilities with an approximate half-maximal degradation concentration (DC_{50}) of 80 nM in an LNCaP cell line. Specificity tests also indicated that compound 1 exhibited a strong preference for LNCaP PrCa, suggesting its potential for targeted therapy in AR-expressing PrCa [188]. In another study Takwale et al. [189] developed various CRBN-mediated AR PROTACs by combining the AR antagonist with the CRBN ligand. Compound TD-802 showed the highest degradation efficiency of AR in LNCaP cells [189]. Targeting AR using RU59063 derivatives, Liang and colleagues [190] developed novel AR PROTACs. Among these AR PROTACs, compound A16 (also known as compound 3) emerged as the leading candidate, matching the efficacy of enzalutamide [190]. Kim and colleagues [191] synthesized PROTACs using bicalutamide analogs with a PEG linker attached to thalidomide, which serves as a CRBN recruiter. Among these PROTACs, compound 13b (compound 4) effectively decreased AR-regulated gene expression. Compound 13c (compound 5) demonstrated even greater AR degradation, particularly in targeting AR-V7, making it more effective than the wild-type AR and suggesting its potential for treating AR-V-expressing PrCAs [191]. In another significant development, Wang et al. [192] developed ARD-61 (compound 14), a highly potent AR degrader. To address the poor oral bioavailability,

Table 1 | Current controllable PROTACs.

Caging strategy	PROTAC name	Target protein	E3 ligand	Reference
Photo-controlled	pc-PROTAC 1	dBET1	CRBN	[176]
	Opto-pomalidomide	IKZF1/3	CRBN	[177-180]
	Opto-dBET1	BRD4	CRBN	
	Opto-dALK	ALK	CRBN	
	Trans-photoPROTAC-1	BRD2	VHL	[181]
	DEACM	ERR α	VHL	[179]
	NPOM	BRD4	CRBN	[179]
	DMNB	BRD4	VHL	[180]
Folate-controlled	folate-PROTAC	BRD3/4	VHL	[182, 183]
Hypoxia-controlled	ha-PROTAC	EGFR	CRBN	[184]
	NTR-PROTAC	EGFR	VHL	[185]
X-ray-controlled	RT-PROTACs	BRD4	VHL	[186]
Stimuli-responsive	sr-PROTAC	BRD4	CRBN	[187]

Table 2 | Summary of AR-PROTAC.

E3	PROTAC name	Reference
CRBN	ARV-110	[8]
	PROTAC 35	[188]
	TD-802	[189]
	A16	[190]
	Compound 13b/13c	[191]
	ARD-2128	[192]
	ARD-2585	[193]
	BWA-522	[200]
	ARD-2051	[201]
	ARD-1676	[202]
VHL	ARD-266	[194]
	A031	[195]
	Niclo-Click PROTAC 5	[196]
	MTX-23	[197]
	ARCC-4	[198]
	ARD-69	[137]
	ARD-61	[199]

Wang et al. [192] optimized the molecule by incorporating the CRBN ligand, resulting in the development of ARD-2128 (compound 6). This compound exhibited excellent oral bioavailability and showed potent AR degradation, significantly reducing AR, PSA, TMPRSS2, and FKBP5 levels, demonstrating the potential as an orally administered AR-targeted therapy [192]. Finally, Xiang et al. [193] developed ARD-2585 (compound 7), which emerged as an efficient degrader of AR. Pharmacokinetic and tissue distribution studies confirmed ARD-2585 promise as an AR degrader, suggesting that ARD-2585 could become a highly effective therapy for PrCa, particularly for patients with resistance to conventional AR inhibitors [193].

In addition to CRBN-recruiting PROTACs, VHL-recruiting PROTACs have also been developed to target AR for degradation with several novel compounds demonstrating promising potential as therapeutic agents in PrCa treatment. Han et al. [194] analysed the correlation between the VHL ligand binding affinity and degradation efficacy. The findings advocated using ligand with a relatively low binding affinity (2–3 μ M) could still produce highly effective AR degraders. Optimized ARD-266 (compound 8) emerged as a stand-out candidate, achieving > 90% AR degradation in 10 nM after further optimization. ARD-266 degraded > 95% AR in PrCa within 6 h [194]. Chen and colleagues [195] reported another degrader, A031 (compound

9), which was optimized from two AR antagonists and several E3 ligands. By comparing the linker composition with different heterocyclic and phenyl rings, Chen et al. [195] reported that the AR ligand rather than the linker has a substantial effect on the overall activity of the PROTACs. Munoz et al. [196] developed niclosamide-based PROTACs that utilized VHL-032 as the VHL ligand. One compound, Niclo-Click PROTAC 5, achieved a half-maximal inhibitory concentration (IC_{50}) of approximately 1 μ M in the LNCaP cell line but the effectiveness was lower than clinical candidates, like ARV-110 [196].

Lee et al. [197] introduced MTX-23 (compound 11), a unique PROTAC that has been demonstrated to degrade both AR-FL and AR-V7. Compound 11 The compound 11 DC_{50} for AR-V7 and AR-FL was 0.37 and 2 μ M in immunoblotting studies, respectively. Compound 11 effectively inhibits cell division and induces programmed cell death in PrCa cells, even in models resistant to enzalutamide and abiraterone [197]. Salami et al. [198] synthesized VHL-based PROTACs targeting AR by utilizing enzalutamide. The most potent compound, ARCC-4 (compound 12), displayed a DC_{50} of 5 nM. ARCC-4 effectively degraded AR in VCaP cells and degraded several clinically relevant AR mutants, demonstrating enhanced efficacy in inducing apoptosis and inhibiting proliferation in CRPC cells [198].

In further optimization efforts, Han et al. [137] developed ARD-69 (compound 13), which exhibits exceptional potency. ARD-69 was optimized by adjusting the linker length and incorporating a pyridine group, enhancing both activity and solubility [137]. Finally, Kregel et al. [199] reported ARD-61 (compound 14), an AR degrader effective against PrCa resistant to standard treatments. ARD-61 triggers PARP cleavage in multiple AR-driven cell lines and exhibits potent activity in the enzalutamide resistant xenograft model. Despite not directly degrading AR-V7, ARD-61 inhibits tumor growth in AR-V7 overexpressing models, suggesting that full-length AR remains crucial for survival in CRPC [199].

Zhang et al. [200] developed a novel PROTAC (BWA-522), which is designed to target to the N-terminal transcriptional domain of AR (AR-NTD). By binding to this domain, BWA-522 induces degradation of AR-FL and the AR-V7 variant in PrCa cell lines. BWA-522 exhibits promising therapeutic value in *in vitro* and *in vivo* studies, with a sub-micromolar DC_{50} value in VCaP cells and a 76% inhibition of tumor growth in LNCaP xenograft mice studies, respectively [200].

ARD-2051, another potent PROTAC degrader of AR, was developed by Han et. al in 2023 [201]. ARD-2051 exhibits a 0.6 nM DC_{50} and effectively inhibits AR-regulated genes, thereby suppressing cancer cell growth. Moreover, oral bioavailability studies indicate favorable pharmacokinetic properties of ARD-2051, making ARD-2051 a potential agent for PrCa [201].

Moreover, ARD-1676, which was developed by Xiang et al. [202], exhibits highly efficient AR degradation,

Review Article

with DC_{50} values of 0.1 and 1.1 nM in VCaP and LNCaP cells, respectively.

In conclusion, the advances in AR-targeted PROTACs provide novel strategies to overcome the limitations of inhibitors, such as enzalutamide, paving the way for enhanced treatment strategies for PrCa.

5.4 Recent developments in non-AR targets

While initially developed to degrade the AR in PrCa due to its critical role in disease progression, the application of PROTACs has been broadened to include non-AR proteins implicated in PrCa, such as BET [203, 204], BCL6 [205], FAK [206], and CDKs [207].

BET proteins, which are key epigenetic regulators, have a central role in gene transcription [208]. The degradation of BET proteins has been shown to inhibit PrCa cell growth, motivating the development of PROTACs targeting BET proteins. For example, the degrader, MZ1, selectively degrades BRD4 by disrupting its interaction with acetylated chromatin [203]. This disruption suppresses MYC-driven gene transcription, ultimately inhibiting cancer cell proliferation. Furthermore, MZ1 has demonstrated potent anti-proliferative effects in prostate cancer models [203].

PROTACs targeting B-cell lymphoma 6 (BCL6), a transcriptional repressor, have been primarily investigated in lymphomas [205]. For example, the PROTAC, BI-3802, induces the proteasomal degradation of BCL6, leading to reactivation of tumor-suppressor genes. While BI-3802 has primarily been studied in lymphomas, the mechanism of action offers valuable insight into the potential for targeting BCL6 pathways in PrCa.

Focal adhesion kinase (FAK), a non-receptor tyrosine kinase involved in cell adhesion and survival signaling pathways, is specifically degraded by PROTACs (PROTAC-FAK-01) [206]. In prostate cancer models, PROTAC-FAK-01 disrupts focal adhesion complexes, effectively suppressing cell migration and proliferation.

Cyclin-dependent kinases (CDKs), such as CDK4/6, serve as key regulators of the cell cycle [209]. PROTACs targeting specific CDKs have been investigated to induce cancer cell apoptosis [207]. Jiang et al. [207] developed dTAG-CDK4/6, a PROTAC that degrades these kinases, with significant tumor-suppressive activity observed in prostate cancer xenograft models.

These developments highlight the versatility of PROTAC technology in targeting a broad range of proteins beyond AR in PrCa, potentially addressing various oncogenic pathways involved in the disease.

5.5 Recent clinical progress of PROTAC technology

Table 3 presents an overview of recent clinical progress in PROTAC technology, highlighting key details, such as disease indications, developmental phases, preliminary outcomes, and the corresponding challenges and implications [8, 107, 141, 142, 200-202].

6. PERSPECTIVE AND FUTURE DIRECTIONS

This section explores the key mechanisms driving AR signaling in CRPC and discusses the future directions for PrCa treatment by harnessing the PROTAC strategy. We focused on the mechanisms underlying AR amplification and overexpression, AR splice variants, mutations leading to receptor promiscuity, post-translational modifications, interactions with co-activators and co-repressors, and intra-tumoral steroid hormone synthesis. Figure 5 highlights the diverse AR-dependent pathways, showcasing therapeutic targets that could potentially be leveraged to enhance CRPC treatment strategies.

6.1 AR amplification and overexpression

AR amplification, characterized by an increased AR gene count surpassing the normal diploid level, is recognized as a prominent characteristic of CRPC and has been identified in 20–31% of cases [210-212]. Fluorescence *in situ* hybridization analysis has shown that AR amplification is uncommon in the early-stage of PrCa but becomes more prevalent as the disease advances to castration-resistant stages, correlating with increased AR mRNA expression. For example, a two-fold increase in AR mRNA levels has been demonstrated in CRPC tumors, highlighting the essential function of AR amplification in enabling tumor adaptation to androgen-depleted conditions [212, 213]. Chen et al. [214] further validated that CRPC cells maintain elevated AR protein levels, emphasizing the function of AR amplification and overexpression in sustaining AR signalling, even in low androgen conditions. In response to these findings, degrader-based therapies have emerged as promising strategies for addressing CRPC driven by AR amplification and overexpression. A notable example is ARV-110, a PROTAC targeting AR, has shown potential in clinical trials by degrading AR while mitigating the effects of overactive AR signalling pathways in CRPC [8].

6.2 Posttranslational modifications

Moreover, post-translational modifications showcase a pivotal role in the stabilization of AR protein in CRPC, thereby contributing to receptor sustained activity. These modifications, such as phosphorylation, acetylation, ubiquitination, and SUMOylation, are crucial regulators that modulate AR stability, promote AR translocation into the nucleus, and fine-tuning AR gene expression and function [215]. Collectively, these post-translational changes ensure prolonged AR signalling, further complicating the management of CRPC and highlighting the urgent need for targeted therapeutic strategies [215]. The interaction with HSPs, particularly the HSP family, acts as a critical molecular chaperone, safeguarding AR from degradation while preserving the active conformation [216]. This protective mechanism allows PrCa cells to remain highly responsive to even minimal levels of circulating hormones, enabling

Table 3 | Summary of clinical progress involving PROTACs and inhibitors.

PROTAC	Target	Indication	Phase of Development	Preliminary Outcomes	Challenges and Implications
Enzalutamide [102]	AR	mCRPC	FDA approved	Improved overall survival and radiographic progression-free survival in phase 3 trials; PREVAIL trial showed a median survival of 32.4 months vs. 30.2 months (placebo).	A standard treatment across prostate cancer stages; resistance development and side effects (fatigue, hypertension, and seizures) limit long-term efficacy.
Bavdegalutamide (ARV-110) [8, 107]	AR	mCRPC	Phase III	Antitumor activity observed, particularly in patients with AR ligand-binding domain mutations (T878 and H875). Demonstrated a manageable tolerability profile in clinical trials.	Companion diagnostics remain essential for identifying patients harboring specific AR mutations. Addressing resistance mechanisms and ensuring therapeutic specificity are critical for success.
NX-5948 [141]	BTk	B-cell malignancies and autoimmune diseases	Phase I	Responses observed within 8 weeks; treatment was well-tolerated with common adverse events including purpura, thrombocytopenia, and neutropenia.	Ensuring selective BTk degradation is vital to minimize off-target effects. Determining optimal dosing regimens and managing immune-related adverse events remain key challenges.
KT-474 [142]	IRAK4	Autoimmune and inflammatory diseases	Phase I	Greater than 95% reduction in IRAK4 levels and up to 97% reduction in pro-inflammatory cytokines observed in healthy volunteers. Maintained a favorable safety profile, with only mild-to-moderate adverse events reported.	Translating findings from healthy volunteers to patients with autoimmune diseases poses challenges. Long-term studies are needed to confirm safety and efficacy for therapeutic potential.
BWA-522 [200]	AR-NTD	Prostate cancer	Preclinical	Demonstrated efficacy in degrading AR-FL and AR-V7, inducing apoptosis in cell lines and achieving significant tumor growth inhibition in xenograft models.	Offers a strategy for targeting resistant AR variants. Challenges include ensuring the translation of preclinical success into clinical efficacy and overcoming resistance mechanisms.
ARD-2051 [201]	AR	Advanced prostate cancer	Preclinical	High degradation potency with $D_{max} > 90\%$, suppression of AR-regulated genes, and inhibition of cancer cell growth were observed in preclinical models. Demonstrated good oral bioavailability	Developing companion diagnostics for AR mutations and refining oral dosing strategies is critical for therapeutic benefit in patients.
ARD-1676 [202]	AR and AR mutants	AR-positive prostate cancer	Preclinical	DC_{50} values ranging from 0.1–1.1 nM in cell lines achieved >90% degradation. Oral administration effectively reduced AR protein levels in tumor tissues and inhibited tumor growth without toxicity.	Exceptional pharmacokinetics and activity against AR mutants suggest significant therapeutic potential. Addressing off-target effects and achieving robust clinical trial outcomes remain key hurdles.

Review Article

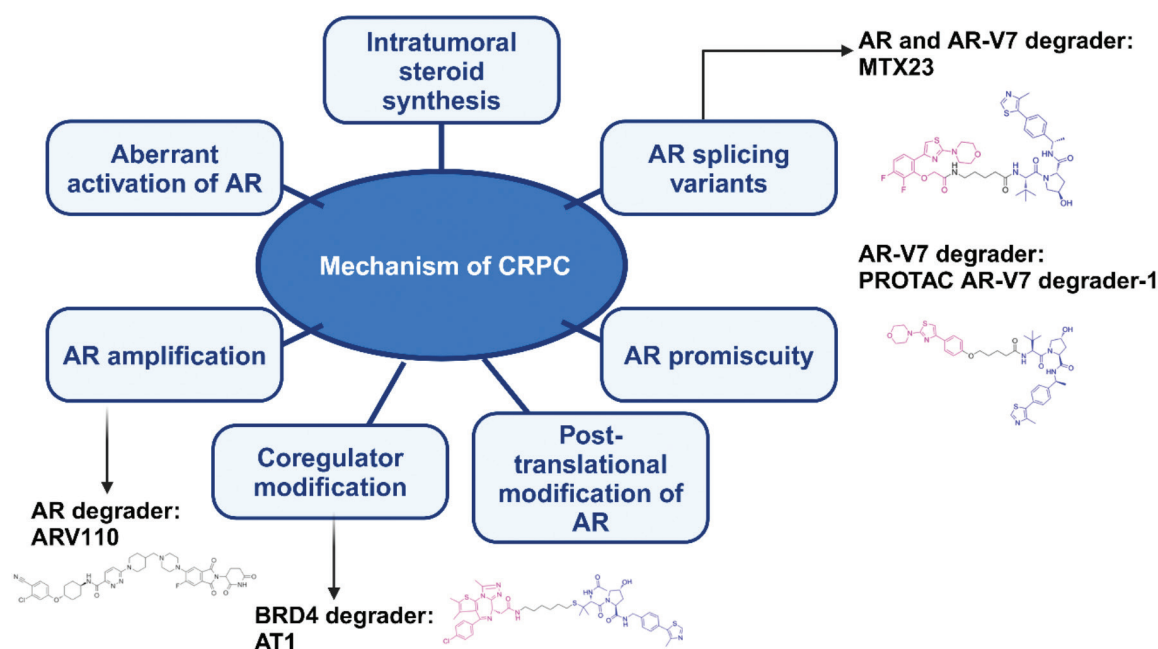


Figure 5 | Potential molecular mechanisms underlying CRPC.

The scheme illustrates seven potential mechanisms of CRPC, including aberrant activation of AR, AR amplification, intra-tumoral steroid synthesis, AR splicing variants, AR promiscuity, post-translational modifications of AR, and coregulator modification.

sustained proliferative growth in an androgen-deficient environment. Consequently, the involvement of HSPs in maintaining AR stability further exacerbates the challenge of targeting AR signaling in CRPC, necessitating more advanced therapeutic approaches to disrupt this protective interaction.

6.3 AR splice variants

Another critical adaptation mechanism in CRPC is the emergence of AR splice variants, which generate truncated AR protein. Among these, AR-V1 and AR-V7 are particularly notable for their roles in therapy resistance. Lacking the LBD but retaining the N-terminal and DNA-binding domains, AR-V1 and V7 function as constitutively active transcription factors independent of androgen binding. This ligand-independent activity enables the splice variants to drive AR signaling continuously, contributing to therapeutic resistance and tumor progression in CRPC [217, 218]. This ligand-independent activity enables AR splice variants to drive downstream pathways even without circulating hormones. The expression has a particularly significant role in driving resistance to enzalutamide and abiraterone [219, 220]. These therapies aim to decrease the activity of AR by inhibiting the LBD domain. However, the AR variants circumvent this blockade because the AR variants lack the LBD. Consequently, AR splice variants contribute to primary resistance, which occurs in 20%–40% of patients, and secondary resistance, which almost invariably develops over time in nearly all treated patients. This finding

highlights the pressing need for therapeutic strategies that effectively target these variants in CRPC [221–223]. This resistance mechanism underscores the urgent need for therapeutic strategies capable of targeting AR splice variants or the downstream effectors to overcome treatment failures in CRPC. Recently, the development of degraders (Figure 5) that target AR and AR-V7 [197, 223] has opened promising new avenues for CRPC treatment, offering a broader range of therapeutic options. As these AR-V7-targeting strategies continue to advance, the strategies warrant increased focus and exploration within the PrCa research field because the strategies hold considerable potential for addressing resistance mechanisms.

In summary, AR amplification, stabilization, splice variants, and other factors have been shown to be critical in maintaining persistent AR signaling in CRPC, underscoring critical pathways that can be exploited for the development of future therapies. A deeper study of these regulatory mechanisms will pave the way for more precise and potent PROTAC strategies, tailored to target these distinct alterations in AR signaling (Figure 5). These advances offer the potential for enhanced therapeutic efficacy and better treatment to patients with PrCa.

ACKNOWLEDGEMENTS

Due to space constraints, we regretfully could not cite all relevant studies and we apologize to our colleagues whose work was omitted in this paper. We are grateful to the members of the Wei laboratory for valuable feedback and

discussions during the review of this manuscript. We thank the US National Institutes of Health (NIH) for supporting this work through grant CA177910 awarded to W.W.

CONFLICTS OF INTEREST

The authors declared that they have no conflicts of interest.

REFERENCES

- [1] Denmeade SR, Isaacs JT: A History of Prostate Cancer Treatment. *Nature Reviews. Cancer* 2002, 2:389–396.
- [2] Wang Z, Wu S, Zhu C, Shen J: The Role of Ferroptosis in Esophageal Cancer. *Cancer Cell International* 2022, 22:266.
- [3] Siegel RL, Miller KD, Fuchs HE, Jemal A: Cancer Statistics, 2022. *CA: A Cancer Journal for Clinicians* 2022, 72:7–33.
- [4] Gandhi J, Afridi A, Vatsia S, Joshi G, Joshi G, Kaplan SA, et al.: The Molecular Biology of Prostate Cancer: Current Understanding and Clinical Implications. *Prostate Cancer and Prostatic Diseases* 2018, 21:22–36.
- [5] Kessel K, Seifert R, Weckesser M, Roll W, Humbert V, Schlack K, et al.: Molecular Analysis of Circulating Tumor Cells of Metastatic Castration-resistant Prostate Cancer Patients receiving (177)Lu-PSMA-617 Radioligand Therapy. *Theranostics* 2020, 10:7645–7655.
- [6] Liang J, Wang L, Poluben L, Nouri M, Arai S, Xie L, et al.: Androgen Receptor Splice Variant 7 Functions Independently of the Full Length Receptor in Prostate Cancer Cells. *Cancer Letters* 2021, 519:172–184.
- [7] Yap TA, Smith AD, Ferraldeschi R, Al-Lazikani B, Workman P, de Bono JS: Drug Discovery in Advanced Prostate Cancer: Translating Biology into Therapy. *Nature Reviews. Drug Discovery* 2016, 15:699–718.
- [8] Bekes M, Langley DR, Crews CM: PROTAC Targeted Protein Degraders: the Past is Prologue. *Nature Reviews Drug Discovery* 2022, 21:181–200.
- [9] He B, Kemppainen JA, Wilson EM: FXXLF and WXXLF Sequences Mediate the NH₂-terminal Interaction with the Ligand Binding Domain of the Androgen Receptor. *The Journal of Biological Chemistry* 2000, 275:22986–22994.
- [10] Loy CJ, Sim KS, Yong EL: Filamin-A Fragment Localizes to the Nucleus to Regulate Androgen Receptor and Coactivator Functions. *Proceedings of the National Academy of Sciences of the United States of America* 2003, 100:4562–4567.
- [11] He B, Kemppainen JA, Voegel JJ, Gronemeyer H, Wilson EM: Activation Function 2 in the Human Androgen Receptor Ligand Binding Domain Mediates Interdomain Communication with the NH₂-Terminal Domain. *The Journal of Biological Chemistry* 1999, 274:37219–37225.
- [12] Veldscholte J, Berrevoets CA, Zegers ND, van der Kwast TH, Grootegoed JA, Mulder E: Hormone-induced Dissociation of the Androgen Receptor-heat-shock Protein Complex: Use of a New Monoclonal Antibody to Distinguish Transformed from Nontransformed Receptors. *Biochemistry* 1992, 31:7422–7430.
- [13] Ozanne DM, Brady ME, Cook S, Gaughan L, Neal DE, Robson CN: Androgen Receptor Nuclear Translocation is Facilitated by the f-actin Cross-linking Protein Filamin. *Molecular Endocrinology* 2000, 14:1618–1626.
- [14] Cardozo CP, Michaud C, Ost MC, Fliss AE, Yang E, Patterson C, et al.: C-Terminal Hsp-interacting Protein Slows Androgen Receptor Synthesis and Reduces Its Rate of Degradation. *Archives of Biochemistry and Biophysics* 2003, 410:134–140.
- [15] Shatkina L, Mink S, Rogatsch H, Klocker H, Langer G, Nestl A, et al.: The Cochaperone Bag-1L Enhances Androgen Receptor Action via Interaction with the NH₂-terminal Region of the Receptor. *Molecular and Cellular Biology* 2003, 23:7189–7197.
- [16] Liao G, Chen LY, Zhang A, Godavorthy A, Xia F, Ghosh JC, et al.: Regulation of Androgen Receptor Activity by the Nuclear Receptor Corepressor SMRT. *The Journal of Biological Chemistry* 2003, 278:5052–5061.
- [17] Cutress ML, Whitaker HC, Mills IG, Stewart M, Neal DE: Structural Basis for the Nuclear Import of the Human Androgen Receptor. *Journal of Cell Science* 2008, 121:957–968.
- [18] He B, Lee LW, Minges JT, Wilson EM: Dependence of Selective Gene Activation on the Androgen Receptor NH₂- and COOH-terminal Interaction. *The Journal of Biological Chemistry* 2002, 277:25631–25639.
- [19] Lee YF, Shyr CR, Thin TH, Lin WJ, Chang C: Convergence of Two Repressors Through Heterodimer Formation of Androgen Receptor and Testicular Orphan Receptor-4: A Unique Signaling Pathway in the Steroid Receptor Superfamily. *Proceedings of the National Academy of Sciences of the United States of America* 1999, 96:14724–14729.
- [20] Rahman M, Miyamoto H, Chang C: Androgen Receptor Coregulators in Prostate Cancer: Mechanisms and Clinical Implications. *Clinical Cancer Research: An Official Journal of the American Association for Cancer Research* 2004, 10:2208–2219.
- [21] Schaufele F, Carbonell X, Guerbodot M, Borngraeber S, Chapman MS, Ma AA, et al.: The Structural Basis of Androgen Receptor Activation: Intramolecular and Intermolecular Amino-carboxy Interactions. *Proceedings of the National Academy of Sciences of the United States of America* 2005, 102:9802–9807.
- [22] Zhou ZX, Sar M, Simental JA, Lane MV, Wilson EM: A Ligand-dependent Bipartite Nuclear Targeting Signal in the Human Androgen Receptor. Requirement for the DNA-binding Domain and Modulation by NH₂-terminal and Carboxyl-terminal Sequences. *The Journal of Biological Chemistry* 1994, 269:13115–13123.
- [23] Cao X, Qin J, Xie Y, Khan O, Dowd F, Scofield M, et al.: Regulator of G-protein Signaling 2 (RGS2) Inhibits Androgen-independent Activation of Androgen Receptor in Prostate Cancer Cells. *Oncogene* 2006, 25:3719–3734.
- [24] Manin M, Baron S, Goossens K, Beaudoin C, Jean C, Veyssiere G, et al.: Androgen Receptor Expression is Regulated by the Phosphoinositide 3-kinase/Akt Pathway in Normal and Tumoral Epithelial Cells. *The Biochemical Journal* 2002, 366:729–736.
- [25] Scheid MP, Woodgett JR: PKB/AKT: Functional Insights from Genetic Models. *Nature Reviews. Molecular Cell Biology* 2001, 2:760–768.
- [26] Yeh S, Lin HK, Kang HY, Thin TH, Lin MF, Chang C: From HER2/Neu Signal Cascade to Androgen Receptor and Its Coactivators: A Novel Pathway by Induction of Androgen Target Genes Through MAP Kinase in Prostate Cancer Cells. *Proceedings of the National Academy of Sciences of the United States of America* 1999, 96:5458–5463.

Review Article

- [27] Gioeli D, Ficarro SB, Kwiek JJ, Aaronson D, Hancock M, Catling AD, et al.: Androgen Receptor Phosphorylation. Regulation and Identification of the Phosphorylation Sites. *The Journal of Biological Chemistry* 2002, 277:29304–29314.
- [28] Blok LJ, de Ruiter PE, Brinkmann AO: Forskolin-induced Dephosphorylation of the Androgen Receptor Impairs Ligand Binding. *Biochemistry* 1998, 37:3850–3857.
- [29] Rochette-Egly C: Nuclear Receptors: Integration of Multiple Signalling Pathways Through Phosphorylation. *Cellular Signalling* 2003, 15:355–366.
- [30] van Royen ME, Cunha SM, Brink MC, Mattern KA, Nigg AL, Dubbink HJ, et al.: Compartmentalization of Androgen Receptor Protein-protein Interactions in Living Cells. *The Journal of Cell Biology* 2007, 177:63–72.
- [31] Heinlein CA, Chang C: Androgen Receptor (AR) Coregulators: An Overview. *Endocrine Reviews* 2002, 23:175–200.
- [32] Powell SM, Christiaens V, Voulgaraki D, Waxman J, Claessens F, Bevan CL: Mechanisms of Androgen Receptor Signalling via Steroid Receptor Coactivator-1 in Prostate. *Endocrine-Related Cancer* 2004, 11:117–130.
- [33] Farla P, Hersmus R, Trapman J, Houtsmuller AB: Antiandrogens Prevent Stable DNA-binding of the Androgen Receptor. *Journal of Cell Science* 2005, 118:4187–4198.
- [34] Klokke TI, Kurys P, Elbi C, Nagaich AK, Hendarwanto A, Slagsvold T, et al.: Ligand-specific Dynamics of the Androgen Receptor at Its Response Element in Living Cells. *Molecular and Cellular Biology* 2007, 27:1823–1843.
- [35] Gaughan L, Logan IR, Neal DE, Robson CN: Regulation of Androgen Receptor and Histone Deacetylase 1 by Mdm2-mediated Ubiquitylation. *Nucleic Acids Research* 2005, 33:13–26.
- [36] Haelens A, Tanner T, Denayer S, Callewaert L, Claessens F: The Hinge Region Regulates DNA Binding, Nuclear Translocation, and Transactivation of the Androgen Receptor. *Cancer Research* 2007, 67:4514–4523.
- [37] Lee DK, Chang C: Endocrine Mechanisms of Disease: Expression and Degradation of Androgen Receptor: Mechanism and Clinical Implication. *The Journal of Clinical Endocrinology and Metabolism* 2003, 88:4043–4054.
- [38] Sheflin L, Keegan B, Zhang W, Spaulding SW: Inhibiting Proteasomes in Human HepG2 and LNCaP Cells Increases Endogenous Androgen Receptor Levels. *Biochemical and Biophysical Research Communications* 2000, 276:144–150.
- [39] Silver DA, Pellicer I, Fair WR, Heston WD, Cordon-Cardo C: Prostate-specific Membrane Antigen Expression in Normal and Malignant Human Tissues. *Clinical Cancer Research: An Official Journal of the American Association for Cancer Research* 1997, 3:81–85.
- [40] Wright GL Jr, Haley C, Beckett ML, Schellhammer PF: Expression of Prostate-specific Membrane Antigen in Normal, Benign, and Malignant Prostate Tissues. *Urologic Oncology* 1995, 1:18–28.
- [41] Rahn KA, Slusher BS, Kaplin AI: Glutamate in CNS Neurodegeneration and Cognition and Its Regulation by GCPII Inhibition. *Current Medicinal Chemistry* 2012, 19:1335–1345.
- [42] Rahn KA, Watkins CC, Alt J, Rais R, Stathis M, Grishkan I, et al.: Inhibition of Glutamate Carboxypeptidase II (GCPII) Activity as a Treatment for Cognitive Impairment in Multiple Sclerosis. *Proceedings of the National Academy of Sciences of the United States of America* 2012, 109:20101–20106.
- [43] Davis MI, Bennett MJ, Thomas LM, Bjorkman PJ: Crystal Structure of Prostate-specific Membrane Antigen, A Tumor Marker and Peptidase. *Proceedings of the National Academy of Sciences of the United States of America* 2005, 102:5981–5986.
- [44] Schulke N, Varlamova OA, Donovan GP, Ma D, Gardner JP, Morrissey DM, et al.: The Homodimer of Prostate-specific Membrane Antigen is a Functional Target for Cancer Therapy. *Proceedings of the National Academy of Sciences of the United States of America* 2003, 100:12590–12595.
- [45] Rawlings ND, Barrett AJ: Structure of Membrane Glutamate Carboxypeptidase. *Biochimica et Biophysica Acta* 1997, 1339:247–252.
- [46] Bernstein LH, Gutstein S, Weiner S, Efron G: The Absorption and Malabsorption of Folic Acid and Its Polyglutamates. *The American Journal of Medicine* 1970, 48:570–579.
- [47] Zhao R, Diop-Bove N, Visentin M, Goldman ID: Mechanisms of Membrane Transport of Folates into Cells and Across Epithelia. *Annual Review of Nutrition* 2011, 31:177–201.
- [48] Figueiredo JC, Grau MV, Haile RW, Sandler RS, Summers RW, Bresalier RS, et al.: Folic Acid and Risk of Prostate Cancer: Results from a Randomized Clinical Trial. *Journal of the National Cancer Institute* 2009, 101:432–435.
- [49] Sanderson SM, Gao X, Dai Z, Locasale JW: Methionine Metabolism in Health and Cancer: A Nexus of Diet and Precision Medicine. *Nature Reviews. Cancer* 2019, 19:625–637.
- [50] Yao V, Bacich DJ: Prostate Specific Membrane Antigen (PSMA) Expression Gives Prostate Cancer Cells a Growth Advantage in a Physiologically Relevant Folate Environment In Vitro. *The Prostate* 2006, 66:867–875.
- [51] Corbin JM, Ruiz-Echevarria MJ: One-Carbon Metabolism in Prostate Cancer: The Role of Androgen Signaling. *International Journal of Molecular Sciences* 2016, 17:1208.
- [52] Zadra G, Ribeiro CF, Chetta P, Ho Y, Cacciatore S, Gao X, et al.: Inhibition of De Novo Lipogenesis Targets Androgen Receptor Signaling in Castration-resistant Prostate Cancer. *Proceedings of the National Academy of Sciences of the United States of America* 2019, 116:631–640.
- [53] Bakht MK, Beltran H: Biological Determinants of PSMA Expression, Regulation and Heterogeneity in Prostate Cancer. *Nature Reviews Urology* 2024, 22:26–45.
- [54] Neale JH, Bzdega T, Wroblewska B: N-Acetylaspartylglutamate: The Most Abundant Peptide Neurotransmitter in the Mammalian Central Nervous System. *Journal of Neurochemistry* 2000, 75:443–452.
- [55] Morland C, Nordengen K: N-Acetyl-Aspartyl-Glutamate in Brain Health and Disease. *International Journal of Molecular Sciences* 2022, 23:1268.
- [56] Kaittanis C, Andreou C, Hieronymus H, Mao N, Foss CA, Eiber M, et al.: Prostate-specific Membrane Antigen Cleavage of Vitamin B9 Stimulates Oncogenic Signaling Through Metabotropic Glutamate Receptors. *The Journal of Experimental Medicine* 2018, 215:159–175.
- [57] Palamiuc L, Emerling BM: PSMA Brings New Flavors to PI3K Signaling: A Role for Glutamate in Prostate Cancer. *The Journal of Experimental Medicine* 2018, 215:17–19.
- [58] Bakht MK, Hayward JJ, Shahbazi-Raz F, Skubal M, Tamura R, Stringer KF, et al.: Identification of Alternative Protein Targets of Glutamate-ureido-lysine Associated with PSMA Tracer Uptake in Prostate Cancer Cells. *Proceedings of the National Academy of Sciences of the United States of America* 2022, 119:e2025710119. <https://doi.org/10.1073/pnas.2025710119>.

- [59] Pellegrino S, Fonti R: A Look into the Future: The Role of PSMA Beyond Prostate Cancer. *European Journal of Nuclear Medicine and Molecular Imaging* 2023, 51:278–280.
- [60] Lee MS, Asomaning K, Su L, Wain JC, Mark EJ, Christiani DC: MTHFR Polymorphisms, Folate Intake and Carcinogen DNA Adducts in the Lung. *International Journal of Cancer* 2012, 131:1203–1209.
- [61] Yao V, Berkman CE, Choi JK, O’Keefe DS, Bacich DJ: Expression of Prostate-specific Membrane Antigen (PSMA), Increases Cell Folate Uptake and Proliferation and Suggests a Novel Role for PSMA in the Uptake of the Non-polyglutamated Folate, Folic Acid. *The Prostate* 2010, 70:305–316.
- [62] Asangani IA, Dommeti VL, Wang X, Malik R, Cieslik M, Yang R, et al.: Therapeutic Targeting of BET Bromodomain Proteins in Castration-resistant Prostate Cancer. *Nature* 2014, 510:278–282.
- [63] Nagarajan S, Hossan T, Alawi M, Najafova Z, Indenbirken D, Bedi U, et al.: Bromodomain Protein BRD4 is Required for Estrogen Receptor-dependent Enhancer Activation and Gene Transcription. *Cell Reports* 2014, 8:460–469.
- [64] Shi J, Vakoc CR: The Mechanisms Behind the Therapeutic Activity of BET Bromodomain Inhibition. *Molecular Cell* 2014, 54:728–736.
- [65] Delmore JE, Issa GC, Lemieux ME, Rahl PB, Shi J, Jacobs HM, et al.: BET Bromodomain Inhibition as a Therapeutic Strategy to Target c-Myc. *Cell* 2011, 146:904–917.
- [66] Wyce A, Degenhardt Y, Bai Y, Le B, Korenchuk S, Crouthame MC, et al.: Inhibition of BET Bromodomain Proteins as a Therapeutic Approach in Prostate Cancer. *Oncotarget* 2013, 4:2419–2429.
- [67] Urbanucci A, Barfeld SJ, Kytola V, Itkonen HM, Coleman IM, Vodak D, et al.: Androgen Receptor Deregulation Drives Bromodomain-Mediated Chromatin Alterations in Prostate Cancer. *Cell Reports* 2017, 19:2045–2059.
- [68] Romero Otero J, Garcia Gomez B, Campos Juanatey F, Touijer KA: Prostate Cancer Biomarkers: An Update. *Urologic Oncology* 2014, 32:252–260.
- [69] Lilja H: Testing New PSA Subforms to Enhance the Accuracy of Predicting Cancer Risk and Disease Outcome in Prostate Cancer. *Clinical Chemistry* 2008, 54:1248–1249.
- [70] Lilja H, Ulmert D, Vickers AJ: Prostate-specific Antigen and Prostate Cancer: Prediction, Detection and Monitoring. *Nature Reviews. Cancer* 2008, 8:268–278.
- [71] Cary KC, Cooperberg MR: Biomarkers in Prostate Cancer Surveillance and Screening: Past, Present, and Future. *Therapeutic Advances in Urology* 2013, 5:318–329.
- [72] Farha MW, Salami SS: Biomarkers for Prostate Cancer Detection and Risk Stratification. *Therapeutic Advances in Urology* 2022, 14:17562872221103988.
- [73] Sokoll LJ, Sanda MG, Feng Z, Kagan J, Mizrahi IA, Broyles DL, et al.: A Prospective, Multicenter, National Cancer Institute Early Detection Research Network study of [-2]proPSA: Improving Prostate Cancer Detection and Correlating with Cancer Aggressiveness. *Cancer Epidemiology, Biomarkers & Prevention: A Publication of the American Association for Cancer Research, Cosponsored by the American Society of Preventive Oncology* 2010, 19:1193–1200.
- [74] Bussemakers MJ, van Bokhoven A, Verhaegh GW, Smit FP, Karthaus HF, Schalken JA, et al.: DD3: A New Prostate-specific Gene, Highly Overexpressed in Prostate Cancer. *Cancer Research* 1999, 59:5975–5979.
- [75] de Kok JB, Verhaegh GW, Roelofs RW, Hessels D, Kiemeny LA, Aalders TW, et al.: DD3(PCA3), A Very Sensitive and Specific Marker to Detect Prostate Tumors. *Cancer Research* 2002, 62:2695–2698.
- [76] Squire JA, Park PC, Yoshimoto M, Alami J, Williams JL, Evans A, et al.: Prostate Cancer as a Model System for Genetic Diversity in Tumors. *Advances in Cancer Research* 2011, 112:183–216.
- [77] Auprich M, Bjartell A, Chun FK, de la Taille A, Freedland SJ, Haese A, et al.: Contemporary Role of Prostate Cancer Antigen 3 in the Management of Prostate Cancer. *European Urology* 2011, 60:1045–1054.
- [78] Schoenborn JR, Nelson P, Fang M: Genomic Profiling Defines Subtypes of Prostate Cancer with the Potential for Therapeutic Stratification. *Clinical Cancer Research: An Official Journal of the American Association for Cancer Research* 2013, 19:4058–4066.
- [79] Crawford ED, Rove KO, Trabulsi EJ, Qian J, Drewnowska KP, Kaminetsky JC, et al.: Diagnostic Performance of PCA3 to Detect Prostate Cancer in Men with Increased Prostate Specific Antigen: A Prospective Study of 1,962 Cases. *The Journal of Urology* 2012, 188:1726–1731.
- [80] Vlaeminck-Guillem V, Ruffion A, Andre J, Devonec M, Paparel P: Urinary Prostate Cancer 3 Test: Toward the Age of Reason? *Urology* 2010, 75:447–453.
- [81] Kupelian PA, Buchsbaum JC, Elshaikh M, Reddy CA, Zippe C, Klein EA: Factors Affecting Recurrence Rates after Prostatectomy or Radiotherapy in Localized Prostate Carcinoma Patients with Biopsy Gleason Score 8 or Above. *Cancer* 2002, 95:2302–2307.
- [82] Sekhoacha M, Riet K, Motloung P, Gumenku L, Adegoke A, Mashele S: Prostate Cancer Review: Genetics, Diagnosis, Treatment Options, and Alternative Approaches. *Molecules* 2022, 27:5730.
- [83] Parker CC, James ND, Brawley CD, Clarke NW, Hoyle AP, Ali A, et al.: Radiotherapy to the Primary Tumour for Newly Diagnosed, Metastatic Prostate Cancer (STAMPEDE): A Randomised Controlled Phase 3 Trial. *Lancet* 2018, 392:2353–2366.
- [84] Sweeney CJ, Chen YH, Carducci M, Liu G, Jarrard DF, Eisenberger M, et al.: Chemohormonal Therapy in Metastatic Hormone-Sensitive Prostate Cancer. *The New England Journal of Medicine* 2015, 373:737–746.
- [85] Grewal K, Grewal K, Tabbara IA: PARP Inhibitors in Prostate Cancer. *Anticancer Research* 2021, 41:551–556.
- [86] Urbanucci A, Mills IG: Bromodomain-containing Proteins in Prostate Cancer. *Molecular and Cellular Endocrinology* 2018, 462:31–40.
- [87] Jones K, Zhang Y, Kong Y, Farah E, Wang R, Li C, et al.: Epigenetics in Prostate Cancer Treatment. *Journal of Translational Genetics and Genomics* 2021, 5:341–356.
- [88] Yan Y, Ma J, Wang D, Lin D, Pang X, Wang S, et al.: The Novel BET-CBP/p300 Dual Inhibitor NEO2734 is Active in SPOP Mutant and Wild-type Prostate Cancer. *MBO Molecular Medicine* 2019, 11:e10659.
- [89] He Y, Wei T, Ye Z, Orme JJ, Lin D, Sheng H, et al.: A Noncanonical AR Addiction Drives Enzalutamide Resistance in Prostate Cancer. *Nature Communications* 2021, 12:1521.
- [90] Sun R, Yan B, Li H, Ding D, Wang L, Pang J, et al.: Androgen Receptor Variants Confer Castration Resistance in Prostate Cancer by Counteracting Antiandrogen-Induced Ferroptosis. *Cancer Research* 2023, 83: 3192–3204.

Review Article

- [91] Paller CJ, Piana D, Eshleman JR, Riel S, Denmeade SR, Isaacsson Velho P, et al.: A Pilot Study of Prostate-specific Membrane Antigen (PSMA) Dynamics in Men Undergoing Treatment for Advanced Prostate Cancer. *The Prostate* 2019, 79:1597–1603.
- [92] Sartor O, de Bono J, Chi KN, Fizazi K, Herrmann K, Rahbar K, et al.: Lutetium-177-PSMA-617 for Metastatic Castration-Resistant Prostate Cancer. *The New England Journal Medicine* 2021, 385:1091–1103.
- [93] Pathmanandavel S, Crumbaker M, Yam AO, Nguyen A, Rofo C, Hovey E, et al.: ¹⁷⁷Lu-PSMA-617 and Idronoxil in Men with End-Stage Metastatic Castration-Resistant Prostate Cancer (LuPIN): Patient Outcomes and Predictors of Treatment Response in a Phase I/II Trial. *Journal of Nuclear Medicine: Official Publication, Society of Nuclear Medicine* 2022, 63:560–566.
- [94] Aggarwal R, Starzinski S, de Kouchkovsky I, Koshkin V, Bose R, Chou J, et al.: Single-dose ¹⁷⁷Lu-PSMA-617 Followed by Maintenance Pembrolizumab in Patients with Metastatic Castration-resistant Prostate Cancer: An Open-label, Dose-expansion, Phase 1 Trial. *The Lancet. Oncology* 2023, 24:1266–1276.
- [95] Ramnarain B, Sartor O: PSMA-Targeted Radiopharmaceuticals in Prostate Cancer: Current Data and New Trials. *The Oncologist* 2023, 28:392–401.
- [96] Buchbinder EI, Desai A: CTLA-4 and PD-1 Pathways: Similarities, Differences, and Implications of Their Inhibition. *American Journal of Clinical Oncology* 2016, 39:98–106.
- [97] Boudadi K, Suzman DL, Anagnostou V, Fu W, Luber B, Wang H, et al.: Ipilimumab Plus Nivolumab and DNA-repair Defects in AR-V7-expressing Metastatic Prostate Cancer. *Oncotarget* 2018, 9:28561–28571.
- [98] Shenderov E, Boudadi K, Fu W, Wang H, Sullivan R, Jordan A, et al.: Nivolumab Plus Ipilimumab, With or Without Enzalutamide, in AR-V7-expressing Metastatic Castration-Resistant Prostate Cancer: A Phase-2 Nonrandomized Clinical Trial. *The Prostate* 2021, 81:326–338.
- [99] Sharma P, Pachynski RK, Narayan V, Flechon 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* 2020, 38:489–499.e3.
- [100] Kyriakopoulos CE, Eickhoff JC, Ferrari AC, Schweizer MT, Wargowski E, Olson BM, et al.: Multicenter Phase I Trial of a DNA Vaccine Encoding the Androgen Receptor Ligand-binding Domain (pTVG-AR, MVI-118) in Patients with Metastatic Prostate Cancer. *Clinical Cancer Research: An Official Journal of the American Association for Cancer Research* 2020, 26:5162–5171.
- [101] Antonarakis ES: Current Understanding of Resistance to Abiraterone and Enzalutamide in Advanced Prostate Cancer. *Clinical Advances in Hematology & Oncology* 2016, 14:316–319.
- [102] La J, Wang L, Corrigan JK, Lang D, Lee MH, Do NV, et al.: Abiraterone or Enzalutamide for Patients With Metastatic Castration-Resistant Prostate Cancer. *JAMA Network Open* 2024, 7:e2428444.
- [103] Beer TM, Armstrong AJ, Rathkopf DE, Loriot Y, Sternberg CN, Higano CS, et al.: Enzalutamide in Metastatic Prostate Cancer Before Chemotherapy. *The New England Journal of Medicine* 2014, 371:424–433.
- [104] Ryan CJ, Smith MR, Fizazi K, Saad F, Mulders PF, Sternberg CN, et al.: Abiraterone Acetate Plus Prednisone Versus Placebo Plus Prednisone in Chemotherapy-naïve Men with Metastatic Castration-resistant Prostate Cancer (COU-AA-302): Final Overall Survival Analysis of a Randomised, Double-blind, Placebo-controlled Phase 3 Study. *The Lancet. Oncology* 2015, 16:152–160.
- [105] Quinn DI, Sandler HM, Horvath LG, Goldkorn A, Eastham JA: The Evolution of Chemotherapy for the Treatment of Prostate Cancer. *Annals of Oncology: Official Journal of the European Society for Medical Oncology* 2017, 28:2658–2669.
- [106] Doyle-Lindrud S: Managing Side Effects of the Novel Taxane Cabazitaxel in Castrate-resistant Prostate Cancer. *Clinical Journal of Oncology Nursing* 2012, 16:286–291.
- [107] Yedla P, Babalghith AO, Andra VV, Syed R: PROTACs in the Management of Prostate Cancer. *Molecules* 2023, 28:3698.
- [108] Cooper G, Adams K: *The Cell: A Molecular Approach*. Oxford University Press; 2022.
- [109] Ahn G, Banik SM, Miller CL, Riley NM, Cochran JR, Bertozzi CR: LYTACs That Engage the Asialoglycoprotein Receptor for Targeted Protein Degradation. *Nature Chemical Biology* 2021, 17:937–946.
- [110] Banik SM, Pedram K, Wisnovsky S, Ahn G, Riley NM, Bertozzi CR: Lysosome-targeting Chimaeras for Degradation of Extracellular Proteins. *Nature* 2020, 584:291–297.
- [111] Marei H, Tsai WK, Kee YS, Ruiz K, He J, Cox C, et al.: Antibody Targeting of E3 Ubiquitin Ligases for Receptor Degradation. *Nature* 2022, 610:182–189.
- [112] Pance K, Gramespacher JA, Byrnes JR, Salangsang F, Serrano JC, Cotton AD, et al.: Modular Cytokine Receptor-targeting Chimeras for Targeted Degradation of Cell Surface and Extracellular Proteins. *Nature Biotechnology* 2023, 41:273–281.
- [113] Zhang D, Duque-Jimenez J, Facchinetti F, Brixi G, Rhee K, Feng WW, et al.: Transferrin Receptor Targeting Chimeras for Membrane Protein Degradation. *Nature* 2024. doi: 10.1038/s41586-024-07947-3
- [114] Hershko A, Ciechanover A: The Ubiquitin System. *Annual Review of Biochemistry* 1998, 67:425–479.
- [115] Komander D, Rape M: The Ubiquitin Code. *Annual Review of Biochemistry* 2012, 81:203–229.
- [116] Vogel G: Nobel Prizes. Gold Medal from Cellular Trash. *Science* 2004, 306:400–401.
- [117] Kanayama A, Seth RB, Sun L, Ea CK, Hong M, Shaito A, et al.: TAB2 and TAB3 Activate the NF-kappaB Pathway Through Binding to Polyubiquitin Chains. *Molecular Cell* 2004, 15:535–548.
- [118] Dang F, Nie L, Wei W: Ubiquitin Signaling in Cell Cycle Control and Tumorigenesis. *Cell Death & Differentiation* 2021, 28:427–438.
- [119] Mevissen TET, Komander D: Mechanisms of Deubiquitinase Specificity and Regulation. *Annual Review of Biochemistry* 2017, 86:159–192.
- [120] Guharoy M, Bhowmick P, Sallam M, Tompa P: Tripartite Degrons Confer Diversity and Specificity on Regulated Protein Degradation in the Ubiquitin-proteasome System. *Nature Communication* 2016, 7:10239.
- [121] Liu J, Tokheim C, Lee JD, Gan W, North BJ, Liu XS, et al.: Genetic Fusions Favor Tumorigenesis Through Degron

- Loss in Oncogenes. *Nature Communication* 2021, 12:6704.
- [122] Liu J, Ma J, Liu Y, Xia J, Li Y, Wang ZP, et al.: PROTACs: A Novel Strategy for Cancer Therapy. *Seminars in Cancer Biology* 2020, 67:171–179.
- [123] Takahashi D, Arimoto H: Targeting Selective Autophagy by AUTAC Degraders. *Autophagy* 2020, 16:765–766.
- [124] Takahashi D, Moriyama J, Nakamura T, Miki E, Takahashi E, Sato A, et al.: AUTACs: Cargo-Specific Degraders Using Selective Autophagy. *Molecular Cell* 2019, 76:797–810e10.
- [125] Li Z, Wang C, Wang Z, Zhu C, Li J, Sha T, et al.: Allele-selective lowering of mutant HTT protein by HTT-LC3 linker compounds. *Nature* 2019, 575:203–209.
- [126] Li Z, Zhu C, Ding Y, Fei Y, Lu B: ATTEC: A Potential New Approach to Target Proteinopathies. *Autophagy* 2020, 16:185–187.
- [127] Sakamoto KM, Kim KB, Kumagai A, Mercurio F, Crews CM, Deshaies RJ: Protacs: Chimeric Molecules that Target Proteins to the Skp1-Cullin-F Box Complex for Ubiquitination and Degradation. *Proceedings of the National Academy of Sciences of the United States of America* 2001, 98:8554–8559.
- [128] Bondeson DP, Mares A, Smith IE, Ko E, Campos S, Miah AH, et al.: Catalytic In Vivo Protein Knockdown by Small-molecule PROTACs. *Nature Chemical Biology* 2015, 11:611–617.
- [129] Schneekloth AR, Pucheault M, Tae HS, Crews CM: Targeted Intracellular Protein Degradation Induced by a Small Molecule: En Route to Chemical Proteomics. *Bioorganic & Medicinal Chemistry Letters* 2008, 18:5904–5908.
- [130] Winter GE, Buckley DL, Paulk J, Roberts JM, Souza A, Dhe-Paganon S, et al.: DRUG DEVELOPMENT. Phthalimide Conjugation as a Strategy for In Vivo Target Protein Degradation. *Science* 2015, 348:1376–1381.
- [131] Sekine K, Takubo K, Kikuchi R, Nishimoto M, Kitagawa M, Abe F, et al.: Small Molecules Destabilize cIAP1 by Activating Auto-ubiquitylation. *The Journal of Biological Chemistry* 2008, 283:8961–8968.
- [132] Lee J, Lee Y, Jung YM, Park JH, Yoo HS, Park J: Discovery of E3 Ligase Ligands for Target Protein Degradation. *Molecules* 2022, 27:6515.
- [133] Bricelj A, Steinebach C, Kuchta R, Gütschow M, Sosič I: E3 Ligase Ligands in Successful PROTACs: An Overview of Syntheses and Linker Attachment Points. *Frontiers in Chemistry* 2021, 9:707317.
- [134] Weng G, Shen C, Cao D, Gao J, Dong X, He Q, et al.: PROTAC-DB: An Online Database of PROTACs. *Nucleic Acids Research* 2021, 49:D1381–D1387.
- [135] He S, Dong G, Cheng J, Wu Y, Sheng C: Strategies for Designing Proteolysis Targeting Chimaeras (PROTACs). *Medicinal Research Reviews* 2022, 42:1280–1342.
- [136] Troup RI, Fallan C, Baud MGJ: Current Strategies for the Design of PROTAC Linkers: A Critical Review. *Exploration of Targeted Anti-tumor Therapy* 2020, 1:273–312.
- [137] Han X, Wang C, Qin C, Xiang W, Fernandez-Salas E, Yang CY, et al.: Discovery of ARD-69 as a Highly Potent Proteolysis Targeting Chimera (PROTAC) Degradator of Androgen Receptor (AR) for the Treatment of Prostate Cancer. *Journal of Medicinal Chemistry* 2019, 62:941–964.
- [138] Morgenthaler M, Schweizer E, Hoffmann-Roder A, Benini F, Martin RE, Jaeschke G, et al.: Predicting and Tuning Physicochemical Properties in Lead Optimization: Amine Basicities. *ChemMedChem* 2007, 2:1100–1115.
- [139] Wiberg KB: Substituent Effects on the Acidity of Weak Acids. 1. Bicyclo[2.2.2]octane-1-carboxylic Acids and bicyclo[1.1.1]pentane-1-carboxylic Acids. *Journal of Organic Chemistry* 2002, 67:1613–1617.
- [140] Davis C, Spaller BL, Matouschek A: Mechanisms of Substrate Recognition by the 26S Proteasome. *Current Opinion in Structural Biology* 2021, 67:161–169.
- [141] Eyre TA, Riches JC: The Evolution of Therapies Targeting Bruton Tyrosine Kinase for the Treatment of Chronic Lymphocytic Leukaemia: Future Perspectives. *Cancers (Basel)* 2023, 15:2596.
- [142] Ackerman L, Acloque G, Bacchelli S, Schwartz H, Feinstein BJ, La Stella P, et al.: IRAK4 Degradator in Hidradenitis Suppurativa and Atopic Dermatitis: A Phase 1 Trial. *Nature Medicine* 2023, 29:3127–3136.
- [143] Dobrovolsky D, Wang ES, Morrow S, Leahy C, Faust T, Nowak RP, et al.: Bruton Tyrosine Kinase Degradation as a Therapeutic Strategy for Cancer. *Blood* 2019, 133:952–961.
- [144] Zarrin AA, Bao K, Lupardus P, Vucic D: Kinase Inhibition in Autoimmunity and Inflammation. *Nature Reviews. Drug Discovery* 2021, 20:39–63.
- [145] Schiemer J, Horst R, Meng Y, Montgomery JI, Xu Y, Feng X, et al.: Snapshots and Ensembles of BTK and cIAP1 Protein Degradator Ternary Complexes. *Nature Chemical Biology* 2021, 17:152–160.
- [146] Zorba A, Nguyen C, Xu Y, Starr J, Borzilleri K, Smith J, et al.: Delineating the Role of Cooperativity in the Design of Potent PROTACs for BTK. *Proceedings of the National Academy of Sciences of the United States of America* 2018, 115:E7285–E7292.
- [147] Tinworth CP, Lithgow H, Dittus L, Bassi ZI, Hughes SE, Muelbaier M, et al.: PROTAC-Mediated Degradation of Bruton's Tyrosine Kinase Is Inhibited by Covalent Binding. *ACS Chemical Biology* 2019, 14:342–347.
- [148] Buhimschi AD, Armstrong HA, Toure M, Jaime-Figueroa S, Chen TL, Lehman AM, et al.: Targeting the C481S Ibrutinib-Resistance Mutation in Bruton's Tyrosine Kinase Using PROTAC-Mediated Degradation. *Biochemistry* 2018, 57:3564–3575.
- [149] Sun Y, Zhao X, Ding N, Gao H, Wu Y, Yang Y, et al.: PROTAC-induced BTK Degradation as a Novel Therapy for Mutated BTK C481S Induced Ibrutinib-resistant B-cell Malignancies. *Cell Research* 2018, 28:779–781.
- [150] Goedert M: Tau Protein and Neurodegeneration. *Seminars in Cell & Developmental Biology* 2004, 15:45–49.
- [151] Chang CW, Shao E, Mucke L: Tau: Enabler of Diverse Brain Disorders and Target of Rapidly Evolving Therapeutic Strategies. *Science* 2021, 371:eabb8255.
- [152] DeVos SL, Miller RL, Schoch KM, Holmes BB, Kebodeaux CS, Wegener AJ, et al.: Tau Reduction Prevents Neuronal Loss and Reverses Pathological Tau Deposition and Seeding in Mice with Tauopathy. *Science Translational Medicine* 2017, 9:eaag0481.
- [153] Wegmann S, DeVos SL, Zeitler B, Marlen K, Bennett RE, Perez-Rando M, et al.: Persistent Repression of Tau in the Brain Using Engineered Zinc Finger Protein Transcription Factors. *Science Advances* 2021, 7:eabe1611.
- [154] Lu M, Liu T, Jiao Q, Ji J, Tao M, Liu Y, et al.: Discovery of a Keap1-dependent Peptide PROTAC to Knockdown

Review Article

- Tau by Ubiquitination-proteasome Degradation Pathway. *European Journal of Medicinal Chemistry* 2018, 146:251–259.
- [155] Wang W, Zhou Q, Jiang T, Li S, Ye J, Zheng J, et al.: A Novel Small-molecule PROTAC Selectively Promotes Tau Clearance to Improve Cognitive Functions in Alzheimer-like Models. *Theranostics* 2021, 11:5279–5295.
- [156] Chien DT, Szardenings AK, Bahri S, Walsh JC, Mu F, Xia C, et al.: Early Clinical PET Imaging Results with the Novel PHF-tau Radioligand [F18]-T808. *Journal of Alzheimer's Disease* 2014, 38:171–184.
- [157] Silva MC, Ferguson FM, Cai Q, Donovan KA, Nandi G, Patnaik D, et al.: Targeted Degradation of Aberrant Tau in Frontotemporal Dementia Patient-derived Neuronal Cell Models. *Elife* 2019, 8:e45457.
- [158] Kotzbauer PT, Trojanowski JQ, Lee VM: Lewy Body Pathology in Alzheimer's Disease. *Journal of Molecular Neuroscience* 2001, 17:225–232.
- [159] Qu J, Ren X, Xue F, He Y, Zhang R, Zheng Y, et al.: Specific Knockdown of α -Synuclein by Peptide-Directed Proteasome Degradation Rescued Its Associated Neurotoxicity. *Cell Chemical Biology* 2020, 27:763.
- [160] A Novel Gene Containing a Trinucleotide Repeat that is Expanded and Unstable on Huntington's Disease Chromosomes. The Huntington's Disease Collaborative Research Group. *Cell* 1993, 72:971–983.
- [161] Sap KA, Reits EA: Strategies to Investigate Ubiquitination in Huntington's Disease. *Frontiers in Chemistry* 2020, 8:485.
- [162] Tomoshige S, Nomura S, Ohgane K, Hashimoto Y, Ishikawa M: Discovery of Small Molecules that Induce the Degradation of Huntingtin. *Angewandte Chemie (International ed. in English)* 2017, 56:11530–11533.
- [163] Li W, Deng X, Chen J: RNA-binding Proteins in Regulating mRNA Stability and Translation: Roles and Mechanisms in Cancer. *Seminars in Cancer Biology* 2022, 86:664–677.
- [164] Kelaini S, Chan C, Cornelius VA, Margariti A: RNA-Binding Proteins Hold Key Roles in Function, Dysfunction, and Disease. *Biology (Basel)* 2021, 10:366.
- [165] Vaquerizas JM, Kummerfeld SK, Teichmann SA, Luscombe NM: A Census of Human Transcription Factors: Function, Expression and Evolution. *Nature Reviews. Genetics* 2009, 10:252–263.
- [166] Hagenbuchner J, Ausserlechner MJ: Targeting Transcription Factors by Small Compounds—Current Strategies and Future Implications. *Biochemical Pharmacology* 2016, 107:1–13.
- [167] Henley MJ, Koehler AN: Advances in Targeting 'Undruggable' Transcription Factors with Small Molecules. *Nature Reviews. Drug Discovery* 2021, 20:669–688.
- [168] Samarasinghe KTG, Jaime-Figueroa S, Burgess M, Nalawansa DA, Dai K, Hu Z, et al.: Targeted Degradation of Transcription Factors by TRAFACs: TRANscription Factor TARgeting Chimeras. *Cell Chemical Biology* 2021, 28:648–661.e5.
- [169] Liu J, Chen H, Kaniskan HU, Xie L, Chen X, Jin J, et al.: TF-PROTACs Enable Targeted Degradation of Transcription Factors. *Journal of the American Chemical Society* 2021, 143:8902–8910.
- [170] Shao J, Yan Y, Ding D, Wang D, He Y, Pan Y, et al.: Destruction of DNA-Binding Proteins by Programmable Oligonucleotide PROTAC (O'PROTAC): Effective Targeting of LEF1 and ERG. *Advanced Science (Weinheim, Baden-Wurttemberg, Germany)* 2021, 8:e2102555.
- [171] Patil KM, Chin D, Seah HL, Shi Q, Lim KW, Phan AT: G4-PROTAC: Targeted Degradation of a G-quadruplex Binding Protein. *Chemical Communications* 2021, 57:12816–12819.
- [172] Wang Z, Liu J, Chen H, Qiu X, Xie L, Kaniskan HU, et al.: Telomere Targeting Chimera Enables Targeted Destruction of Telomeric Repeat-Binding Factor Proteins. *Journal of the American Chemical Society* 2023, 145:10872–10879.
- [173] Li X, Zhang Z, Gao F, Ma Y, Wei D, Lu Z, et al.: c-Myc-Targeting PROTAC Based on a TNA-DNA Bivalent Binder for Combination Therapy of Triple-Negative Breast Cancer. *Journal of the American Chemical Society* 2023, 145:9334–9342.
- [174] Wang Z, Liu J, Qiu X, Zhang D, Inuzuka H, Chen L, et al.: Methylated Nucleotide-Based Proteolysis-Targeting Chimera Enables Targeted Degradation of Methyl-CpG-Binding Protein 2. *Journal of the American Chemical Society* 2023, 145:21871–21878.
- [175] Wang Z, Zhang D, Qiu X, Inuzuka H, Xiong Y, Liu J, et al.: Structurally Specific Z-DNA Proteolysis Targeting Chimera Enables Targeted Degradation of Adenosine Deaminase Acting on RNA 1. *Journal of the American Chemical Society* 2024, 146:7584–7593.
- [176] Xue G, Wang K, Zhou D, Zhong H, Pan Z: Light-Induced Protein Degradation with Photocaged PROTACs. *Journal of the American Chemical Society* 2019, 141:18370–18374.
- [177] Liu J, Chen H, Ma L, He Z, Wang D, Liu Y, et al.: Light-induced Control of Protein Destruction by Opto-PROTAC. *Science Advances* 2020, 6:eaay5154.
- [178] Li Z, Ma S, Yang X, Zhang L, Liang D, Dong G, et al.: Development of Photocontrolled BRD4 PROTACs for Tongue Squamous Cell Carcinoma (TSCC). *Europeana Journal of Medicinal Chemistry* 2021, 222:113608.
- [179] Naro Y, Darrah K, Deiters A: Optical Control of Small Molecule-Induced Protein Degradation. *Journal of the American Chemical Society* 2020, 142:2193–2197.
- [180] Kounde CS, Shchepinova MM, Saunders CN, Muelbauer M, Rackham MD, Harling JD, et al.: A Caged E3 Ligase Ligand for PROTAC-mediated Protein Degradation with Light. *Chemical Communications (Cambridge, England)* 2020, 56:5532–5535.
- [181] Pfaff P, Samarasinghe KTG, Crews CM, Carreira EM: Reversible Spatiotemporal Control of Induced Protein Degradation by Bistable PhotoPROTACs. *ACS Central Science* 2019, 5:1682–1690.
- [182] Liu J, Chen H, Liu Y, Shen Y, Meng F, Kaniskan HU, et al.: Cancer Selective Target Degradation by Folate-Caged PROTACs. *Journal of the American Chemical Society* 2021, 143:7380–7387.
- [183] Chen H, Liu J, Kaniskan HU, Wei W, Jin J: Folate-Guided Protein Degradation by Immunomodulatory Imide Drug-Based Molecular Glues and Proteolysis Targeting Chimeras. *Journal of Medicinal Chemistry* 2021, 64:12273–12285.
- [184] Cheng W, Li S, Wen X, Han S, Wang S, Wei H, et al.: Development of Hypoxia-activated PROTAC Exerting a More Potent Effect in Tumor Hypoxia than in Normoxia. *Chemical Communications* 2021, 57:12852–12855.

- [185] Shi S, Du Y, Zou Y, Niu J, Cai Z, Wang X, et al.: Rational Design for Nitroreductase (NTR)-Responsive Proteolysis Targeting Chimeras (PROTACs) Selectively Targeting Tumor Tissues. *Journal of Medicinal Chemistry* 2022, 65:5057–5071.
- [186] Yang C, Yang Y, Li Y, Ni Q, Li J: Radiotherapy-Triggered Proteolysis Targeting Chimera Prodrug Activation in Tumors. *Journal of the American Chemical Society* 2023, 145:385–391.
- [187] An K, Deng X, Chi H, Zhang Y, Li Y, Cheng M, et al.: Stimuli-Responsive PROTACs for Controlled Protein Degradation. *Angewandte Chemie (International ed. in English)* 2023, 62:e202306824.
- [188] Gockel LM, Pfeifer V, Baltes F, Bachmaier RD, Wagner KG, Bendas G, et al.: Design, Synthesis, and Characterization of PROTACs Targeting the Androgen Receptor in Prostate and Lung Cancer Models. *Archiv der Pharmazie (Weinheim)* 2022, 355:e2100467.
- [189] Takwale AD, Jo SH, Jeon YU, Kim HS, Shin CH, Lee HK, et al.: Design and Characterization of Cereblon-mediated Androgen Receptor Proteolysis-targeting Chimeras. *European Journal of Medicinal Chemistry* 2020, 208:112769.
- [190] Liang JJ, Xie H, Yang RH, Wang N, Zheng ZJ, Zhou C, et al.: Designed, Synthesized and Biological Evaluation of Proteolysis Targeting Chimeras (PROTACs) as AR Degraders for Prostate Cancer Treatment. *Bioorganic & Medicinal Chemistry* 2021, 45:116331.
- [191] Kim GY, Song CW, Yang YS, Lee NR, Yoo HS, Son SH, et al.: Chemical Degradation of Androgen Receptor (AR) Using Bicalutamide Analog-Thalidomide PROTACs. *Molecules* 2021, 26:2525.
- [192] Han X, Zhao L, Xiang W, Qin C, Miao B, McEachern D, et al.: Strategies Toward Discovery of Potent and Orally Bioavailable Proteolysis Targeting Chimera Degraders of Androgen Receptor for the Treatment of Prostate Cancer. *Journal of Medicinal Chemistry* 2021, 64:12831–12854.
- [193] Xiang W, Zhao L, Han X, Qin C, Miao B, McEachern D, et al.: Discovery of ARD-2585 as an Exceptionally Potent and Orally Active PROTAC Degradar of Androgen Receptor for the Treatment of Advanced Prostate Cancer. *Journal of Medicinal Chemistry* 2021, 64:13487–13509.
- [194] Han X, Zhao L, Xiang W, Qin C, Miao B, Xu T, et al.: Discovery of Highly Potent and Efficient PROTAC Degraders of Androgen Receptor (AR) by Employing Weak Binding Affinity VHL E3 Ligase Ligands. *Journal of Medicinal Chemistry* 2019, 62:11218–11231.
- [195] Chen L, Han L, Mao S, Xu P, Xu X, Zhao R, et al.: Discovery of A031 as Effective Proteolysis Targeting Chimera (PROTAC) Androgen Receptor (AR) Degradar for the Treatment of Prostate Cancer. *European Journal of Medicinal Chemistry* 2021, 216:113307.
- [196] Munoz E, Chen G, Hossain A, Wu S, Ocegüera Nava E, Hang J, et al.: Synthesis and Biological Evaluation of Niclosamide PROTACs. *Bioorganic & Medicinal Chemistry Letters* 2022, 72:128870.
- [197] Lee GT, Nagaya N, Desantis J, Madura K, Sabaawy HE, Kim WJ, et al.: Effects of MTX-23, A Novel PROTAC of Androgen Receptor Splice Variant-7 and Androgen Receptor, on CRPC Resistant to Second-Line Antiandrogen Therapy. *Molecular Cancer Therapeutics* 2021, 20:490–499.
- [198] Lotem J, Sachs L: Genetic Dissection of the Control of Normal Differentiation in Myeloid Leukemic Cells. *Proceedings of the National Academy of Sciences of the United States of America* 1977, 74:5554–5558.
- [199] Kregel S, Wang C, Han X, Xiao L, Fernandez-Salas E, Bawa P, et al.: Androgen Receptor Degraders Overcome Common Resistance Mechanisms Developed During Prostate Cancer Treatment. *Neoplasia* 2020, 22:111–119.
- [200] Zhang B, Liu C, Yang Z, Zhang S, Hu X, Li B, et al.: Discovery of BWA-522, a First-in-Class and Orally Bioavailable PROTAC Degradar of the Androgen Receptor Targeting N-Terminal Domain for the Treatment of Prostate Cancer. *Journal of Medicinal Chemistry* 2023, 66:11158–11186.
- [201] Han X, Zhao L, Xiang W, Miao B, Qin C, Wang M, et al.: Discovery of ARD-2051 as a Potent and Orally Efficacious Proteolysis Targeting Chimera (PROTAC) Degradar of Androgen Receptor for the Treatment of Advanced Prostate Cancer. *Journal of Medicinal Chemistry* 2023, 66:8822–8843.
- [202] Xiang W, Zhao L, Han X, Xu T, Kregel S, Wang M, et al.: Discovery of ARD-1676 as a Highly Potent and Orally Efficacious AR PROTAC Degradar with a Broad Activity against AR Mutants for the Treatment of AR + Human Prostate Cancer. *Journal of Medicinal Chemistry* 2023, 66:13280–13303.
- [203] Ma L, Wang J, Zhang Y, Fang F, Ling J, Chu X, et al.: BRD4 PROTAC Degradar MZ1 Exerts Anticancer Effects in Acute Myeloid Leukemia by Targeting c-Myc and ANP32B Genes. *Cancer Biology & Therapy* 2022, 23:1–15.
- [204] Raina K, Lu J, Qian Y, Altieri M, Gordon D, Rossi AM, et al.: PROTAC-induced BET Protein Degradation as a Therapy for Castration-resistant Prostate Cancer. *Proceedings of the National Academy of Sciences of the United States of America* 2016, 113:7124–7129.
- [205] Slabicki M, Yoon H, Koepfel J, Nitsch L, Roy Burman SS, Di Genua C, et al.: Small-molecule-induced Polymerization Triggers Degradation of BCL6. *Nature* 2020, 588:164–168.
- [206] Cromm PM, Samarasinghe KTG, Hines J, Crews CM: Addressing Kinase-Independent Functions of Fak via PROTAC-Mediated Degradation. *Journal of the American Chemical Society* 2018, 140:17019–17026.
- [207] Jiang B, Wang ES, Donovan KA, Liang Y, Fischer ES, Zhang T, et al.: Development of Dual and Selective Degraders of Cyclin-Dependent Kinases 4 and 6. *Angewandte Chemie (International ed. in English)* 2019, 58:6321–6326.
- [208] Wang ZQ, Zhang ZC, Wu YY, Pi YN, Lou SH, Liu TB, et al.: Bromodomain and Extraterminal (BET) Proteins: Biological Functions, Diseases, and Targeted Therapy. *Signal Transduction and Targeted Therapy* 2023, 8:420.
- [209] Sherr CJ, Beach D, Shapiro GI: Targeting CDK4 and CDK6: From Discovery to Therapy. *Cancer Discovery* 2016, 6:353–367.
- [210] Visakorpi T, Hyytinen E, Koivisto P, Tanner M, Keinänen R, Palmberg C, et al.: In Vivo Amplification of the Androgen Receptor Gene and Progression of Human Prostate Cancer. *Nature Genetics* 1995, 9:401–406.
- [211] Bubendorf L, Kononen J, Koivisto P, Schraml P, Moch H, Gasser TC, et al.: Survey of Gene Amplifications During Prostate Cancer Progression by High-throughput Fluorescence In Situ Hybridization on Tissue Microarrays. *Cancer Research* 1999, 59:803–806.
- [212] Linja MJ, Savinainen KJ, Saramaki OR, Tammela TL, Vessella RL, Visakorpi T: Amplification and Overexpression

Review Article

- of Androgen Receptor Gene in Hormone-refractory Prostate Cancer. *Cancer Research* 2001, 61:3550–3555.
- [213] Wadosky KM, Koochekpour S: Molecular Mechanisms Underlying Resistance to Androgen Deprivation Therapy in Prostate Cancer. *Oncotarget* 2016, 7:64447–64470.
- [214] Chen CD, Welsbie DS, Tran C, Baek SH, Chen R, Vessella R, et al.: Molecular Determinants of Resistance to Antiandrogen Therapy. *Nature Medicine* 2004, 10:33–39.
- [215] van der Steen T, Tindall DJ, Huang H: Posttranslational modification of the androgen receptor in prostate cancer. *International Journal of Molecular Sciences* 2013, 14:14833–14859.
- [216] Azad AA, Zoubeidi A, Gleave ME, Chi KN: Targeting Heat Shock Proteins in Metastatic Castration-resistant Prostate Cancer. *Nature Reviews Urology* 2015, 12:26–36.
- [217] Hu R, Dunn TA, Wei S, Isharwal S, Veltri RW, Humphreys E, et al.: Ligand-independent Androgen Receptor Variants Derived from Splicing of Cryptic Exons Signify Hormone-refractory Prostate Cancer. *Cancer Research* 2009, 69:16–22.
- [218] Dehm SM, Schmidt LJ, Heemers HV, Vessella RL, Tindall DJ: Splicing of a Novel Androgen Receptor Exon Generates a Constitutively Active Androgen Receptor that Mediates Prostate Cancer Therapy Resistance. *Cancer Research* 2008, 68:5469–5477.
- [219] Nakazawa M, Antonarakis ES, Luo J: Androgen Receptor Splice Variants in the Era of Enzalutamide and Abiraterone. *Hormones & Cancer* 2014, 5:265–273.
- [220] Antonarakis ES, Lu C, Wang H, Luber B, Nakazawa M, Roeser JC, et al.: AR-V7 and Resistance to Enzalutamide and Abiraterone in Prostate Cancer. *The New England Journal of Medicine* 2014, 371:1028–1038.
- [221] Scher HI, Fizazi K, Saad F, Taplin ME, Sternberg CN, Miller K, et al.: Increased Survival with Enzalutamide in Prostate Cancer after Chemotherapy. *The New England Journal of Medicine* 2012, 367:1187–1197.
- [222] de Bono JS, Logothetis CJ, Molina A, Fizazi K, North S, Chu L, et al.: Abiraterone and Increased Survival in Metastatic Prostate Cancer. *The New England Journal of Medicine* 2011, 364:1995–2005.
- [223] Ryan CJ, Smith MR, de Bono JS, Molina A, Logothetis CJ, de Souza P, et al.: Abiraterone in Metastatic Prostate Cancer Without Previous Chemotherapy. *The New England Journal of Medicine* 2013, 368:138–148.