**The disruption of protein-protein interactions as a therapeutic strategy for prostate cancer**

Bárbara Matos[1], John Howl[2], Carmen Jerónimo[3,4], Margarida Fardilha[1]

[1] Laboratory of Signal Transduction, Department of Medical Sciences, Institute of Biomedicine – iBiMED, University of Aveiro, 3810-193 Aveiro, Portugal

[2] Molecular Pharmacology Group, Research Institute in Healthcare Science, University of Wolverhampton, Wolverhampton WV1 1LY, UK

[3] Cancer Biology and Epigenetics Group, IPO Porto Research Center (CI-IPOP), Portuguese Institute of Oncology of Porto (IPO Porto), Research Center-LAB 3, F Bdg., 1st floor, Rua Dr. António Bernardino de Almeida, 4200-072 Porto, Portugal

[4] Department of Pathology and Molecular Immunology, Institute of Biomedical Sciences Abel Salazar– University of Porto (ICBAS-UP), Porto, Portugal

**Corresponding author:**

Margarida Fardilha, PhD

Department of Medical Sciences, Institute of Biomedicine – iBiMED, University of Aveiro, 3810-193 Aveiro, Portugal

Tel.: +351234247240

e-mail: mfardilha@ua.pt

**Abstract**

Prostate cancer (PCa) is one of the most common male-specific cancers worldwide, with high morbidity and mortality rates associated with advanced disease stages. The current treatment options of PCa are prostatectomy, hormonal therapy, chemotherapy or radiotherapy, the selection of which is usually dependent upon the stage of the disease. The development of PCa to a castration-resistant phenotype (CRPC) is associated with a more severe prognosis requiring the development of a new and effective therapy. Protein-protein interactions (PPIs) have been recognised as an emerging drug modality and targeting PPIs is a promising therapeutic approach for several diseases, including cancer. The efficacy of several compounds in which target PPIs and consequently impair disease progression were validated in phase I/II clinical trials for different types of cancer. In PCa, various small molecules and peptides proved successful in inhibiting important PPIs, mainly associated with the androgen receptor (AR), Bcl-2 family proteins, and kinases/phosphatases, thus impairing the growth of PCa cells *in vitro*. Moreover, a majority of these compounds require further validation *in vivo* and, preferably, in clinical trials. In addition, several other PPIs associated with PCa progression have been identified and now require experimental validation as potential therapeutic loci.

In conclusion, we consider the disruption of PPIs to be a promising though challenging therapeutic strategy for PCa. Agents which modulate PPIs might be employed as a monotherapy or as an adjunct to classical chemotherapeutics to overcome drug resistance and improve efficacy. The discovery of new PPIs with important roles in disease progression, and of novel optimized strategies to target them, are major challenges for the scientific and pharmacological communities.

**Keywords:** protein-protein interactions ; disruption ; prostate cancer ; treatment ; small molecules ; peptides

Abbreviations: AR: androgen receptor; Bcl-2: B-cell lymphoma 2; CRPC: castration-resistant prostate cancer; HSPs: heat shock proteins; HTS: high throughput screening; PAINS: pan-assay interference compounds; PCa: prostate cancer; PPIs: protein-protein interactions; PSA: prostate-specific antigen

## 1. Introduction

Prostate cancer (PCa) is the second most common type of cancer and the fifth leading cause of cancer-associated mortality among men worldwide[1]. The PCa treatment is critically dependent upon the stage of the disease. Frequently, in early-stage and localized disease, the radical prostatectomy or local radiotherapy are the preferred treatment options for PCa. When the tumor has spread outside of the prostate, or if first-line treatments fail, hormone therapy is then employed. Hormone therapy is applied since PCa cells growth are highly dependent on androgens. This type of treatment consists on deplete, often through castration, or block the action of androgens. Overall, these treatment options are associated with a good survival rate[2,3]. Nevertheless, in a non-negligible number of cases, PCa adapts to survive and grow under castration levels of androgens, becoming castration-resistant (CRPC). The molecular mechanisms through which CRPC develops are not fully clarified; however the amplification of androgen receptor (AR), the expression of AR splice variants, an increased interaction of AR with its coactivators and secondary androgen production by PCa cells are likely contributing factors[4,5]. Indeed, the AR is considered a vital driver of CRPC progression and most of the first-line current treatments for CRPC are based on targeting the AR signaling pathway. Besides AR signaling, other pathways seem to be dysregulated in CRPC and have emerged as CRPC drug targets. These alternative targets include PI3K/Akt/mTOR, Wnt/ β-catenin and Hippo signaling pathways[6–8]. The therapeutic manipulation of heat shock proteins (HSPs) and AR co-activators has also been proposed for CRPC treatment. Unfortunately, and despite these many potential therapeutic advancements, the prognosis for CRPC patients remains unsatisfactory and the development of an effective treatment for CRPC remains a major challenge [5,9].

Protein-protein interactions (PPIs) and micro-RNA-target interactions were considered important in deciphering the mechanisms of tumorigenesis and were proposed as potential therapeutic biomarkers[10–12]. Targeting PPIs has emerged as a promising therapeutic approach for several types of cancer. In the last decades, large scale efforts have surveyed and catalogued PPIs which, collectively, constitute the human interactome[13,14]. These studies have identified a large number of PPIs with critical influences upon major signalling pathways. Indeed, though many PPIs lack detailed characterization, their involvement in critical cellular functions, including cell growth and differentiation, DNA replication,

transcriptional activation, translation and transmembrane signal transduction is evident[15,16]. Deregulation of these protein interactions is often associated with pathologic conditions. In fact, more than 650, 000 disease-relevant PPIs have been reported, however about 98% of these interactions remain underexplored[17].

Predominantly because of their highly complex and dynamic nature, PPIs have traditionally been considered "undruggable". Nevertheless, in response to advancements in technological expertise, coupled with increased scientific knowledge, PPIs have emerged as a promising drug targets [14]. Consequently, recent years have witnessed an exponential increase in reports detailing the successful disruption of PPIs, with an emphasis upon diseases of the brain and cardiovascular system [15.16].

In this context, we have rigorously reviewed the potential of PPI disruption as a therapeutic strategy for PCa. Herein, we summarize a range of contemporary approaches and their outcomes. We also describe potential PPIs that, whilst their disruption has not yet been tested, are associated with disease progression. Finally, we highlight the most significant conclusions to be drawn from such studies and elaborate upon future research opportunities in the field.

## 2. Targeting PPIs in cancer

The knowledge of cancer genomics is essential to the molecular characterization of human cancers since it allows the definition of cancer-associated genes and their respective proteins. Recently, attention is turning towards the understanding of how these proteins interact and form PPIs that contribute to dysregulated oncogenic pathways[18]. Despite most of the current PPIs databases do not contain cancer-specific analysis, several attempts have been made to identify cancer-associated PPIs. In addition, dysregulation of various PPIs was associated with cancer initiation and/or progression. First, based upon lung cancer-associated genes libraries, Li *et al* (2017)[19] identified 260 cancer-associated PPIs. Thereafter, Ivanov *et al* (2018)[20] integrated all the available information about PPIs with roles in lung cancer to create an integrative resource named OncoPPI Portal. This platform comprises more than 2500 cancer-associated PPIs. Curiously, about 85% of cancer-associated PPIs are novel, when compared with those described in public databases, indicating that most of them are exclusive to cancer tissues. More recently, a pan-cancer mapping of differential PPIs was

performed by Gulfidan *et al* (2020)[21]. A total of 2039 cancer-associated PPIs were common to eleven of all types of cancer investigated[21]. Nevertheless, the currently available tools for exploring oncogenic PPIs networks are limited and the identification of cancer-related PPIs remains challenging[20].

The important role of PPIs in forming signaling networks that transmit pathophysiological signals to achieve an integrated biological output, allowing the acquisition of hallmark features of cancer, is well established. Indeed, the involvement of PPIs in tumorigenesis, tumor progression, invasion and metastasis have been reported[21,22]. Individually and collectively, these findings suggest that the disruption of PPIs critical for cancer progression could offer a novel and effective therapeutic strategy[12].

Thus, in recent years, PPIs, that have long been considered "undruggable" have developed into attractive molecular targets for novel anticancer therapies. Nevertheless, it has been shown that only about 32% of cancer-associated PPIs are druggable[21]. The development of PPI inhibitors remains a time-consuming, expensive, and difficult process, which should only be initiated for the best validated targets. Indeed, it is estimated that only a small proportion of the proposed compounds is successfully tested in clinical trials[23]. The significant challenges associated with the targeting of PPIs are a consequence of their large interface areas, lack of deep pockets, presence of non-contiguous binding sites, general lack of natural ligands, and the intracellular location of most of them[12]. In general, PPI inhibitors should be able to enter into the cell and mimic the binding hotspots which support discrete protein interactions to ensure high affinity binding; retain some proprieties of the same target to overcome drug resistance; and be active not only to the proposed PPIs, but also their paralogs[24]. These challenges can, however, be overcome so today there are multiple examples of PPI inhibitors with beneficial results for different types of cancer[25–27], including PCa. Most of them have been exclusively analysed i*n vitro*, and pre-clinical and clinical validations are still required to confirm their therapeutic effects.

To the best of our knowledge, to date, 14 small molecule PPI inhibitors have been subjected to clinical trials as possible anticancer therapies. These compounds inhibit PPIs belonging to three main pathways: Bcl family inhibitors[28–34], MDM2-p53 inhibitors[35–40] and SMAC-XIAP inhibitors[41–45]. In Phase I/II clinical trials of gossypol and its R enantiomer, a limited efficacy was demonstrated in breast and prostate cancers[28,29]. Other Bcl2 inhibitors

(Obatoclax, Navitoclax and Venetoclax) were tested in a phase I study and despite some side effects, they were well tolerated and safe. These compounds exhibited considerable clinical activities for lung cancer, lymphoma and acute myelogenous leukemia[30–32,34]. Navitoclax was associated with limited single-agent activity and combination with other drugs was suggested, while Venetoclax had significant antitumor activity as a single-agent, in Phase II clinical trials[33,34]. Several MDM2-p53 inhibitors were also evaluated in Phase I clinical trials. In particular, RG7112, RG7388, DS-3032b, HK-8242 and JNJ-26854165 demonstrated acceptable safety and tolerability, and evidence of clinical activity against solid tumors and lymphoma. Most of them also restored the p53 tumor suppressor activity[35–40]. The SMAC-XIAP inhibitors AT-406, GDC-01542, GDC-0917 and HGS1029 were safe and tolerable for most solid tumors and lymphoma[41–44]. The oral administration of AT-406 was tested in a Phase II clinical trial and limited antitumor activity was observed. This compound was suggested as an adjuvant of classic anticancer therapy[41]. Thus, many PPIs inhibitors tested in clinical trials proved successful either as monotherapy or in combination with classic chemotherapeutic agents, for several types of cancer.

## 3. Evidence acquisition

A relevant bibliography was selected after an extensive Web of Science search up to April 15$^{th}$ 2020, using the keywords: ”protein protein interaction”, “disruption” and “prostate cancer”. Reference lists from the articles were also examined for potentially useful studies. Furthermore, enrolled articles were selected if following the next criteria: written in English, central theme based on disruption of PPIs in PCa, and clinical relevance for PCa treatment.

## 4. Targeting PPIs in PCa: what has been done?

### 4.1. PPIs targeted by small molecules

In recent years, one of the major goals of drug discovery has been the development of small molecules with the ability to target specific PPIs[46]. Despite the challenges associated with this strategy, particularly the extended surface areas of PPI interfaces, several small organic molecules proved successful in blocking PPIs with important roles in disease progression, both *in vitro* and *in vivo*[47]. The main studies in this topic are summarized in the following sections (**Table 1**).

#### 4.1.1. Androgen receptor (AR)-related PPIs

Androgens, acting through the AR, are essential for prostate development and homeostasis. AR signaling has been associated with tumor growth, anti-apoptotic ability and dysregulated lipid pathway in PCa[48,49]. Indeed, AR, as a nuclear receptor, facilitates ligand-dependent transcriptional activation and interacts with several tissue-specific transcriptional factors, inducing the proliferation of prostate epithelial cells and PCa tumor growth[50]. Thus, targeting androgens and AR signaling are considered therapeutic strategies for the majority of prostate tumors[51]. Androgen deprivation therapy is one of the mainstays of PCa treatment, as it can suppress the disease progression. Moreover, targeting the AR axis has also been considered the first-line approach in the treatment of PCa, when it evolves to CRPC, since AR alterations are the main features of this condition. More recently, the disruption of PPIs involving AR with small molecules emerged as a strategy to affect the dysregulated signaling pathways, resulting in a better outcome for PCa patients. This approach has led to some promising results **(Table 1)**.

Since AR transcriptional activity is modulated by coregulatory proteins, it is logical to consider these PPIs as targets for agents designed to decrease AR transactivation[52]. Hence, two small molecule compounds were developed to disrupt interactions of AR with its coactivators. The pterostilbene (PTER)-isothiocyanate (ITC) conjugate was associated with anti-androgenic proprieties, by disrupting AR interaction with its coactivators SRC-1 and GRIP-1, downregulating AR activity (**Figure 1A**). Thus, PTER-ITC caused accumulation of PCa cells in the G2/M phase and induced apoptosis[53]. The disruption of AR interaction with its coactivator β-catenin was also achieved using a small inhibitor of nuclear β-catenin activity, named iCRT3. The disruption of this interaction resulted in inhibition of PCa cell proliferation (**Figure 1A**)[54] and repression of tumor growth in mice xenografts[54]. Moreover, an aberrant activation of AR through Wnt/β-catenin signaling was associated with the progression of PCa to CRPC[5], suggesting that iCRT3 may have also a beneficial effect in CRPC treatment. Nevertheless, this compound was also described as an inhibitor of TCF/β-catenin and thus affects the Wnt pathway. Targeting this signaling pathway has been associated with several limitations. The important role of this signaling pathway in the developmental process and in tissue homeostasis by regulating a wide range of cellular processes, makes it difficult to discretely modulate this pathway without significant side effects. Indeed, and despite substantial efforts from the scientific and pharmacological communities to circumvent these

limitations, no therapeutic agent targeting the Wnt pathway is currently approved for cancer therapy[55].

The AR interacts with several HSPs, contributing to PCa cell survival and proliferation[56]. The beneficial effect of ailanthone in inhibiting tumor growth was demonstrated both *in vitro* and *in vivo* and was associated with the disruption of AR/HSP90 interaction. Indeed, ailanthone, a natural compound recognized as a potent inhibitor of AR, could reduce AR nuclear translocation by targeting the co-chaperone protein p23, and thus preventing the interaction of AR with HSP90 (**Figure 1B**). This small molecule was also associated with decreased CRPC cells proliferation and metastasis[57]. Enzalutamide is an antagonist of AR and is the first therapeutic approach for CRPC. However, a significant percentage of CRPC tumors develop resistance to this drug. Ailanthone also offers advantages in overcoming this drug-resistance associated with many CRPC[57]. The administration of this small molecule in mice xenografts demonstrated a good safety and low toxicity, which highlighted the potential of this drug in CRPC treatment[57]. Furthermore, the interaction of AR with Hsp27 also facilitates AR translocation to the nucleus and its transactivation, thus contributing to PCa cell survival. Apartorsen (OGX-427), an antisense oligonucleotide inhibitor that targets Hsp27, disrupted the AR/Hsp27 interaction to promote the ubiquitin/proteasome-mediated degradation of AR and increase PCa cell apoptosis (**Figure 1B**)[58]. A phase I clinical trial of OGX-427 to treat CRPC demonstrated its good safety and tolerability[59]. Moreover, in a phase II clinical trial, OGX-427 was administered in combination with prednisone, a corticosteroid commonly used in PCa treatment. In this study, CRPC patients had decreased prostate-specific antigen (PSA) levels, though no changes in the proportion of CRPC patients without disease progression were observed after 12 weeks [59].

Other PPIs involving AR have been disrupted. For instance, the cyclin-dependent kinase 5 (Cdk5) has been considered a regulator of AR[60]. Indeed, Cdk5, by interacting with AR, enables its phosphorylation, resulting in the stabilization of AR. The reduction of AR/Cdk5 interaction by roscovitine decreased PCa cells proliferation (**Figure 1C**)[61]. In addition, the combination of this compound with Akt inhibitors was associated with apoptosis of metastatic PCa cells[62]. Nevertheless, roscovitine were considered a pan-CDK inhibitor and thus, its effects on other targets should be carefully analyzed, since they could limit its efficacy.

### 4.1.2. B-cell lymphoma-2 (Bcl-2) protein family-related PPIs

Upregulation of antiapoptotic B-cell lymphoma-2 (Bcl-2) family proteins is observed in almost all human tumors, including PCa, and seems to play key roles in apoptosis resistance, contributing to cancer progression[63–66]. These proteins exert their protective effects by the direct binding and sequestering of their pro-apoptotic counterparts (Bax, Bak and Bad)[67]. The overexpression of Bcl-2 was also observed in the progression of PCa to an androgen-independent stage (CRPC)[68]. Based on these roles in tumor progression, the inhibition of Bcl-2 protein family members represents a novel and promising therapeutic strategy for PCa and CRPC. The potential of Bcl-2 protein family members as therapeutic targets has long been considered, but many putative Bcl-2 inhibitors are non-specific, influencing other cellular targets. Besides, their non-mechanism-based toxicities also limit their effectiveness[67]. Nevertheless, some approaches to target Bcl-2 family-associated protein interactions have been developed (**Table 1**).

The BH3 interacting motif, necessary for the interaction of Bcl-2 with its pro-apoptotic counterparts, is a major target for disruption. Several natural BH3 mimetics have been identified in the last years, including quercetin and gossypol. Quercetin proved successful in disrupting the Bcl-xL/Bax interaction, inducing the release of pro-apoptotic proteins, to trigger apoptosis in PCa cells (**Figure 2A**)[69]. A role in reversing the resistance to docetaxel, a drug used for CRPC treatment for over a decade, was also proposed[70]. Similarly, (-)-gossypol inhibited PCa cell growth and induced apoptosis through mitochondrial pathways by blocking the Bcl-$x_L$/Bax or Bad interactions (**Figure 2A**). Additionally, (-)-gossypol synergistically enhanced the antitumor activity of docetaxel and radiation therapy *in vitro* and *in vivo,* suggesting its combination with other agents in PCa and CRPC treatment[71,72]. Subsequently, this compound was tested in phase I/II clinical trials though a limited efficacy was demonstrated. Despite the decrease in PSA levels observed in some patients, no objective response, according to RECIST guidelines, was observed[29]. Moreover, quercetin and (-)-gossypol were considered pan-assay interference compounds (PAINS), which have emerged as a limitation inherent of many natural products[73]. PAINS are compounds that appear in several high throughput screenings (HTS) against different targets, indicating that they have a wide range of cellular targets, which inhibit their development into successful probes, and makes them associated with several side effects[74]. There are a few examples

when it was possible to omit the “PAIN moieties” without loss of activity. However, this is an underexplored and limited approach and PAINS continue to be considered as false positives in many HTS[73,75].

The fat-soluble vitamin α-tocopheryl succinate was also associated with BH3 mimetic activity. The treatment of PCa cells with this agent induced apoptosis associated with the disruption of the interaction of Bcl-$x_L$ and Bcl-2 with Bak (**Figure 2A**)[76]. This compound, despite its target multiplicity, seems to have a selective action in tumor cells. Indeed, minimal effects were observed in normal cells, however more detailed studies are still required[77]. The Bcl-2 synthetic inhibitor ABT-737 was synthetized using structure-based design with BH3 region of Bad and has been considered a novel anticancer drug[78,79]. Like some natural Bcl-2 inhibitors, ABT-737 inhibited the Bcl-2/Bax and Bcl-$x_L$/Bak interactions, inducing apoptosis (**Figure 2A**)[79]. ABT-737 presented higher affinity than others Bcl-2 inhibitors and its major problem was associated with intrinsic resistance of PCa cells, caused by the expression of anti-apoptotic Mcl-1 and additional apoptosis regulation pathways. This limitation was overcome by combining ABT-737 with other compounds that decrease Mcl-1 expression. In fact, ABT-737, when combined with docetaxel and an immunotoxin, both inhibiting the expression of Mcl-1, improve the anticancer effects of these compounds[79,80]. This agent has also been tested for the treatment of other types of cancer and, in lung cancer, problems in drug delivery arise. To solve this problem, an oral version of ABT-737, named ABT-263, which share binding profile and affinities was developed and successfully tested[81].

The disruption of Bcl-2/Bax was also successful with non BH3 mimetics, including diallyl trisulfide (DATS). Decreased interaction of Bcl-2/Bax, together with Bcl-2 phosphorylation and cleavage of procaspase-9 and-3, were associated with increased apoptosis of PCa cells incubated with DATS (**Figure 2A**)[82]. *In vivo*, the oral administration of this compound resulted in inhibition of tumor progression and pulmonary metastasis[83]. DATS also supressed AR function, which contributed to its effect on PCa progression[84]. This agent was considered promiscuous due to the modulation of multiple signaling pathways. Nevertheless, DATS seems to have higher cytotoxicity in PCa cells, compared with normal prostate cells[85].

Other strategies involving the disruption of Bcl-2 protein family members have also yielded positive outcomes for PCa treatment[86,87]. Bcl-2/Beclin-1 (Becn1) interaction represents a convergence point between apoptosis and autophagy. The interaction with Bcl-2 leads to the

repression of Becn1 pro-autophagic activity[88]. Besides its potential to disrupt PPIs between Bcl-2 family members[71], (-)-gossypol can also interrupt the interaction between Bcl-2 or Bcl-$x_L$ and Becn1, releasing Bcn1, which triggered PCa cells autophagy (**Figure 2B**)[86]. This effect was confirmed *in vivo*, when the oral administration of (-)-gossypol significantly inhibited PCa growth in mice xenografts[86]. Moreover, 3-azido withaferin A (3-AWA), a derivative of the natural product withaferin, has been considered a strong anticancer candidate. This compound was considered a highly selective MMP-2 inhibitor and it seems to be through this effect that 3-AWA affects Bcl-2-associated PPIs. Several other targets have also been described for this compound[89,90] Indeed, Bcl-2/Becn-1 interactions were inhibited in PCa cells incubated with 3-AWA, which sensitized PCa cells to apoptosis (**Figure 2B**)[87].

The targeting of other interactions involving pro-apoptotic Bcl-2 family proteins has been tested using different approaches[91–93]. The Ku70 protein, commonly associated with cancer progression and chemoresistance, has been recognized as a Bax suppressor[94]. Therefore, the natural flavonoid apigenin disrupted the Bax/Ku70 interaction, leading to increased Bax levels and consequently inducing PCa cells apoptosis, *in vitro* and *in vivo* (**Figure 2C**)[91]. The apoptotic effect of apigenin in PCa cells was also mediated by disruption of Bad/ 14-3-3β interaction (**Figure 2D**)[92]. A role in CRPC treatment was suggested for this compound[95]. Nevertheless, apigenin, like quercetin and gossypol was considered a PAINS, the wide range of intracellular protein targets limiting its success in drug development[96,97]. Furthermore, the inactivation of the Akt signaling axis achieved with DATS, resulted in the disruption of Bad/14-3-3β interaction ultimately leading to the apoptosis of PCa cells, and consequent beneficial effect (**Figure 2D**)[93].

#### 4.1.3. Other transcription/ translation factors-related PPIs

Targeting transcription factors to modulate aberrant gene expression in cancer has become a reality[96]. In PCa, several transcription and translation factors were associated with drug resistance, disease progression and metastasis[97]. Besides AR, mentioned in the previous section, PPIs involving other transcription/ translation factors have been blocked using different approaches.

Adaptation to hypoxic microenvironments is a common feature of solid tumors. In PCa, hypoxia can drive disease progression and was associated with increased risk of invasion,

metastasis, treatment failure, and mortality[98,99]. Hypoxia-inducible factor 1 (HIF-1) is a key factor activated in hypoxic conditions to regulate the adaptative response of cancer cells to low oxygen concentrations[100]. Estrogen-related receptor alpha (ERRα), which is overexpressed in PCa, directly interacts with HIF-1α and inhibits its ubiquitination. As a result, the repression of ERRα/HIF-1α interaction by XCT790, a selective and potent ERRα inverse agonist, was associated with attenuation of ERRα-enhanced hypoxic PCa cells growth[101]. Moreover, different epidithiodiketopiperazines (ETPs), including gliotoxin, chaetoxin and chetomin, can also disrupt HIF-1α signaling. Indeed, ETPs blocked the interaction of HIF-1α with its coactivator p300, with consequent decrease of PCa tumor growth and angiogenesis *in vitro* and *in vivo*[102]. Due to its structure, it was suggested that ETPs may have other targets, which should be carefully analyzed in future studies[103].

The ability of ZNF433, which is overexpressed in most prostate tumors, to enhance β-catenin binding to transcription factor 4 (TCF4) is associated with PCa growth and migration, by regulating the expression of target genes[104]. This interaction was antagonized by a diterpenoid derivative - NC043, which directly targets CARF to block β-catenin/ TCF4, decreasing the malignant behaviour of PCa cells[105,106]. An impaired prostate tumorigenesis was also observed when mice xenografts were treated with NC043[106]. As previously mentioned, the Wnt/β -catenin pathway is one of the most significant pathways in CRPC initiation and progression and, thus, the potential of NC043 in CRPC treatment can be suggested[7]. Despite these promising results for NC043, as previously mentioned, targeting the Wnt pathway has been associated with several problems[55]. Furthermore, c-Myc, which is considered a key transcriptional effector of Wnt signaling, also contributes to prostate tumor progression, by promoting PCa cells survival. Thus, targeting c-Myc has been proposed as a potential therapeutic strategy for PCa[107]. , Despite the successfully inhibition of c-Myc/ Max by [Z,E]-5-[4-ethylbenzylidine]-2-thioxothiazolidin-4-one (10058-F4) *in vitro*, no significant inhibition of tumor growth was observed *in vivo* after intravenous treatment with this compound [108,109]. The 10058-F4 compound was associated with a complete specificity for c-Myc/Max interaction and the lack of antitumor activity *in vivo* may have been caused by low concentration in tumor cells and/or by a rapid metabolism[109,110]. Thus, the formulation of this compounds needs optimization. Besides, it has been suggested that c-Myc drives the progression of CRPC, mainly by increasing the expression and activity of AR and AR

splice variants. Indeed, the inhibition of c-Myc sensitized enzalutamide-resistant PCa cells to growth inhibition[111]. Thus, 10058-F4 seems to have a potential role in CRPC treatment.

Defects of the central translation process have also begun to be considered an important contributor to PCa development. Thus, targeting translation has become a novel approach for PCa treatment[112]. The repression of PPIs involving translation initiation factors has been associated with decreased tumor resistance to chemotherapy and castration[113,114]. The phenazine#14 compound, a natural compound that selectively disrupted the interaction between eukaryotic translation initiation factor 4E (eIF4E) and Hsp27, decreased the viability of chemo- and castration-resistant PCa cells[113]. This effect was confirmed *in vitro* and *in vivo*[113]. Similarly, *in vitro* incubation of PCa cells with dactolisib (BEZ235), a PI3K/mTOR inhibitor, disrupted eIF4E/eIF4G interaction, affecting cells chemoresistance[114]. The effect of this compound in PI3K/mTOR signaling suggests a potential role in CRPC treatment.

#### 4.1.4. Kinases/ phosphatases-related PPIs

Because of their important roles in the vast majority of signal transduction processes, the potential of protein kinases and phosphatases as therapeutic targets has rightly been considered[115]. Protein kinases and phosphatases are often dysregulated in pathological conditions, including cancer[116]. Several protein kinases have been implicated in PCa cell survival and proliferation. Indeed, decreased PCa cell growth was observed after the specific knockdown of several protein kinases[117]. Some protein phosphatases have also been associated with PCa cell growth, differentiation, survival, and metastatic potential[118,119]. In particular, the blockage of various PPIs involving protein kinases and/or phosphatases has been associated with improved PCa outcomes[120–123].

The interaction between protein tyrosine kinase 2 beta (PYK2) and receptor tyrosine-protein kinase erbB-2 (ErbB-2) was associated with the adhesive ability of PCa cells and ERK/MAPK activity is a mediator of this effect[120]. Both the expression of a PYK2 mutant protein (K457A—PYK2) and the treatment of cells with a synthetic, potent and selective MEK inhibitor - PD98059 decreased PCa cell adhesion capacity by abolishing the PYK2/ErbB-2 interaction (**Figure 3A**)[120]. This compound also enhanced the docetaxel-induced apoptosis of CRPC cells, suggesting a beneficial role in the treatment of CRPC [124]. Moreover, the natural carbazole alkaloid mahanine, decreased PCa cell survival and proliferation by inhibiting the

interaction between Akt and DNA methyltransferases 1 (DNMT1) and 3B (DNMT3B)[121]. The repression of Akt/DNMTs interactions induced proteasomal degradation of DNMTs, with consequent demethylation of Ras-association domain family 1A (RASSF1A), restoring its expression and tumor suppressor activity (**Figure 3B**)[121]. Mahanine also disrupted AR signaling by inhibiting androgen-dependent and-independent transactivation, suggesting a potential role in CRPC treatment [125]. Despite these beneficial effects, mahanine was associated with a polypharmacological action, modulating multiple kinases, a feature that may limit mahanine-based drug development[126].

PCa has been associated with a dramatic decrease in intracellular zinc levels, compared with benign prostate tissue[127]. Through cooperation with tumor suppressor p53, zinc repressed the interaction between HK2 and VDAC1, by activating GSK3β, resulting in PCa cell apoptosis[122]. This effect was confirmed *in vivo*, using a mice xenograft model (**Figure 3C**)[122].

The protein phosphatase 2 (PP2A) has been implicated in the impairment of PCa cell growth. Indeed, decreased levels of PP2A were associated with increased PCa cell survival, growth and migration[128]. This protein associates with midline-1 (MID1) and the regulatory α4, forming a complex that mediates PP2A degradation[123]. By disrupting the PP2A/MID1/α4 complex, metformin increased PP2A activity to inhibit PCa cell growth and migration and reduce AR protein levels, suggesting beneficial effects to be used in PCa treatment (**Figure 3D**)[123]. Metformin has been described for the treatment of different pathologic conditions and is the most commonly prescribed treatment for Type 2 Diabetes[129]. More recently, a role in the treatment of different types of cancer has been proposed. This compound can interact with several metabolites and hormones and targets multiple signaling pathways with key roles in cancer initiation and progression[130]. Despite its target multiplicity, metformin was associated with a favourable toxicity profile with moderate side effects. A phase II clinical trial demonstrated a role of metformin in CRPC treatment, by inducing disease stabilization[131].

#### 4.1.5. Other PPIs

The disruption of additional PPIs are suggestive of alternative approaches for PCa treatment[132,133]. For instance, the targeting of Poly [ADP-ribose] polymerase 2 (PARP2), which is overexpressed in PCa, has been recognized as an efficient approach to inhibit AR

signaling, since PARP2 is a critical component of AR transcriptional machinery[134,135]. Indeed, blocking PARP2/FOXA1 interaction with 5-(2-oxo-2-phenylethoxy)-1(2H)-isoquinolinone (UPF 1069), a synthetic selective inhibitor of PARP2, attenuated AR-mediated gene expression and inhibited AR-mediated PCa cell growth[132]. Because of the AR-associated mechanism of action, a role of UPF 1069 in CRPC treatment was described[132]. In addition, the tankyrase protein (TNKS), also a member of the PARP family, was considered a crucial mediator of Wnt signal transduction associated with prostate tumorigenesis and disease aggressiveness[136]. TNKS is stabilized by interacting with ubiquitin-specific protease 25 (USP25). The selective repression of this complex was achieved using a small molecule named C44, causing the reduction of PCa cell proliferation both *in vitro* and *in vivo*[133]. Through affecting Wnt signaling, which is crucial for the development of CRPC[7], a role of C44 in CRPC treatment can be hypothesized, however, it is important to consider the previous mentioned limitations associated with targeting Wnt pathway[55].

Increased PCa cells apoptosis was also observed when some PPIs were inhibited[137,138]. The synthetic small molecule inhibitor of apoptosis proteins (IAP) antagonist SH-130 was able to disrupt the interaction between X-linked IAP and Smac[137]. The incubation of PCa cells with SH-130, enhanced the radiation-induced apoptosis. This effect was confirmed *in vivo* using mice xenografts intravenously injected with SH-130. The injected mice overcame apoptosis resistance and the sensibilization to radiotherapy was improved[137]. CRPC was also associated with radiation resistance and IAPs seems to play a key role. Thus, SH-130 was also proposed for the treatment of CRPC. Indeed, IAP antagonists has been associated with a key role in overcome radiation and chemo-therapy resistance. For instance, these compounds increased sensitivity and amplified the apoptotic response to enzalutamide[139].

DNA damage is a mechanism that can promote intrinsic cell apoptosis[140]. PARP inhibitors have been long proposed as anticancer therapies, due to the role of PARP in DNA damage repair. Inhibiting PARP leads to increase DNA damage, resulting in cancer cells apoptosis. Indeed, recently, two PARP inhibitors were approved by FDA for the treatment of PCa and CRPC[141]. The PARP inhibitor veliparib also demonstrated beneficial effects for the treatment of PCa. The E3 ubiquitin-protein ligase UHRF1/ Breast cancer type 1 susceptibility protein (BRCA1) complex, responsible for DNA damage repair, was disrupted by this compound, resulting in PCa cells apoptosis[138]. Recently, the dual PARP/HDAC inhibitor

treatment was suggested for anticancer treatment[142]. In fact, the co-administration of veliparib with the HDAC inhibitor SAHA in mice xenografts synergistically inhibited the prostate tumor growth[138].

Finally, the invasive and metastatic ability of PCa cells were also affected by disrupting different PPIs[143,144]. Simvastatin, one of the most common and extensively researched statins, prevented the interaction of PCa cells with the endothelium. Specifically, simvastatin represses the interaction between integrin $\alpha_v\beta_3$ and endothelial Intercellular Adhesion Molecule 1 (ICAM1), thus inhibiting PCa cell metastasis[143]. A role in delaying CRPC metastasis was also described for simvastatin[145]. Simvastatin is a drug commonly used to lower cholesterol and its main target is an enzyme involved in cholesterol synthesis[146]. This could lead to side effects, however high levels of circulating cholesterol was associated with an increased risk of aggressive PCa, which suggest that the simvastatin-mediated decrease in cholesterol levels is beneficial for PCa patients[147]. This compound is currently being studied in a phase I/II clinical trial for its potential use in PCa treatment (ClinicalTrials.gov Identifier: NCT00572468). Coordination of dynamic microtubules and actin filaments has been implicated in PCa cells invasion[148]. Therefore, the blockage of the drebrin/EB3 complex by 3,5-bis(trifluoromethyl)pyrazole 2 (BTP2), a small molecule inhibitor of drebrin that bind to actin filaments, resulted in the decreased invasive ability of PCa cells[144]. Indeed, several pyrazole derivatives have been associated with anticancer activities and some of them were proposed as potential anticancer drugs[149].

### 4.2. PPIs targeted by peptides

Larger macromolecules, including peptides, have long been proposed to disrupt PPIs. Traditionally, therapeutic peptides were derived from natural sources but, in more recent years, the solid phase synthesis of rationally-designed peptides has revolutionized molecular pharmacology[150]. The emergence of these macromolecules as new PPIs chemical inhibitors has also expanded the repertoire of druggable PPIs. The applications of peptides and peptide-like materials to target PPIs aims to overcome the limitations associated with small molecules. The major advantages of peptides are their reduced immunogenicity, improved safety and high selectivity and potency[151]. Nonetheless, the application of peptides in living cells is often hampered by insufficient cell permeability and proteolytic instability[152]. More recently, these limitations have been overcome and the potential of peptides is growing

rapidly and, to date, there are over 60 peptide drugs approved[153]. The applications of peptides to disrupt several PPIs important for PCa progression are described in following sections (**Table 2**).

#### 4.2.1. Androgen receptor (AR)-related PPIs

Recognition of the AR as the major therapeutic target in PCa has triggered the development of new and improved therapeutic strategies (**Table 2**)[154]. The repression of AR interaction with its co-activators has demonstrated effects in the impairment of PCa progression[155–157]. A synthetic peptide mimicking the structure of SRC-1 was able to selectively disrupt the AR interaction with its co-activators SRC-1 and SRC-2, to decrease PCa cells proliferation (**Figure4A**). In addition to reducing AR and AR variant V-7 transactivation, this peptide also inhibited the AR-dependent expression of PSA in a CRPC cell line, suggesting its utilization in CRPC treatment[155]. The proline-glutamic acid- and leucine-rich protein 1 (PELP1) is also a co-activator of AR and its interaction involves the LXXLL motif. The AR/PELP1 interaction was disrupted by a peptidomimetic, named D2, containing the LXXLL binding motif[156]. D2 blocked the androgen-induced nuclear uptake and genomic activity of AR, with a consequent abrogation of PCa cells proliferation (**Figure 4A**). This beneficial effect was confirmed *in vivo*, using a mouse xenograft model. In addition to these beneficial effects, this compound was considered stable, non-toxic and efficiently taken up by PCa cells, highlighting its potential use in PCa treatment[156]. The PPI between the AR and its coactivator gelsolin (GSN) was also disrupted by mimetic peptides containing either the whole or partial DNA or ligand binding domain. Such peptides blocked the AR/GSN interaction resulting in suppression of GSN-enhanced AR activity (**Figure 4A**)[157]. However, the authors highlighted the possible disruption of other interactions of AR with its regulators using these peptides, since the region covered by the peptides is common to the interaction with several AR regulators. These effects needs to be carefully analyzed[157].

Other AR interacting proteins seem to play a role in PCa pathogenesis. By disrupting the interaction of AR with SWI/SNF-related matrix-associated actin-dependent regulator of chromatin subfamily E member 1 (BAF57), the BIPep, a $NH_2$-terminal inhibitory peptide of BAF57, antagonized AR function and, consequently, decreased PCa cells proliferation (**Figure 4B**)[158]. In addition, PCa cells proliferation was also strongly impaired by a peptide that targeted the SH3 binding motif. This motif mediates the interaction of AR with the proto-

oncogene tyrosine-protein kinase Src (**Figure 4C**). The intraperitoneal injection of the same peptide into a mouse xenograft suppressed the tumor growth, corroborating a beneficial effect in PCa treatment[159]. Finally, the serine/threonine-protein kinase 4 (MST1) is considered a negative regulator of the interaction between AR and Yes-associated protein 1 (YAP1). YAP1 is considered a key effector of Hippo pathway and, by interacting with AR conferred castration-resistance to PCa[8]. Thus, MST1 seems to play a role in CRPC treatment. Indeed, the repression of AR/YAP1 interaction by MST1 decreased the proliferation of invasive CRPC cells **(Figure 4D**)[160].

#### 4.2.2. Kinases/ phosphatases-related PPIs

The blockage of PPIs involving protein kinases or phosphatases, with the ultimate goal of impairing PCa progression, has also been achieved using peptides[161–164]. As kinases and phosphatases often have docking sites outside their active site, selective peptides can be engineered to mimic these unique PPI interfaces. Thus, it is possible for peptides to disrupt kinase/phosphatase-related PPIs without compromising the efficient catalysis of the active site[150]. The nonapeptide KRX-123 disrupted the interaction between Lyn kinase and its substrates by targeting a unique interaction site within Lyn, resulting in decreased PCa cells proliferation (**Figure 5A**)[161]. The PCa tumor regression was observed after intravenous injection of KRX-123 in a PCa mice xenograft model, corroborating the *in vitro* results. Lyn was considered a prime target for CRPC, which led authors to suggest KRX-123 for CRPC treatment [161]. Moreover, the destabilization of Nm23-H1/h-Prune interaction by a competitive permeable peptide (CPP) resulted in increased PCa cells apoptosis *in vitro* and inhibited metastasis *in vivo (***Figure 5B***).* The CPP was also associated with inhibition of Akt/mTOR and NF-kB signaling pathways, which suggests that this peptide could have multiple targets[162]. Lastly, when phosphorylated by Akt, a forkhead box protein O1 (FOXO1)-derived peptide inhibitor bound to Ras GTPase-activating-like protein IQGAP1 (IQGAP1), suppressing the IQGAP1/MAPK interaction. The blockage of this PPI proved to be successful in suppressing the taxane chemoresistance (**Figure 5C**)[163].

The blockage of the signaling of protein phosphatases with peptides has also shown beneficial effects. The interaction of phosphatidylinositol 3,4,5-triphosphate 5-phosphatase 2 (Ship 2) with the Ephrin type-A receptor 2 (EphA2) receptor tyrosine kinase, based on their Sam domains, can be disrupted by the $(KRI)_3$ peptide conjugated with a cell penetrating sequence

and a fluorescent probe – FITC-TAT-$(KRI)_3$ peptide. By damaging the PCa cells plasma membrane, the FITC-TAT-$(KRI)_3$ peptide increased the PCa cells necrosis *in vitro*, contributing to the reduction of tumor growth (**Figure 5D**)[164].

#### 4.2.3. Other PPIs

The disruption of other PPIs, involving different types of proteins with critical roles in PCa progression, has proven successful in the suppression of disease progression. Targeting Rho GTPase guanine nucleotide exchange factor Vav3 (Vav3), which is overexpressed in PCa, has been recognized as an efficient approach to inhibit AR signaling[165]. Indeed, blocking Vav3/cell division cycle 37 homolog (Cdc37) interaction, by using a peptide corresponding to Vav-3 binding region of Cdc37, attenuated AR transcriptional activity and inhibited PCa cells proliferation. A role of this interaction in the progression of PCa to CRPC was proposed by authors, who suggested the potential use of the synthesized peptide in CRPC treatment[166]. More recently, peptide macrocycles (peptoids) have emerged to improve the disruption of PPIs, by mimicking protein secondary structure motifs. A peptoid that binds to a pocket in the interface between TCF and β-catenin was able to suppress this interaction, inhibiting Wnt and AR signaling, with consequent suppression of cells proliferation[167]. The inhibition of Wnt signaling was confirmed *in vivo* using a zebrafish model[167]. The effect of the synthesized peptoid in Wnt signaling suggested a potential role in CRPC treatment. Nevertheless, as mentioned in previous section, problems with targeting Wnt signaling could emerge[55].

The interaction of CC chemokine receptor-9 (CCR9) with its natural ligand CCL25 was associated with the upregulation of anti-apoptotic proteins and inhibition of cytotoxic effects of etoposide, an anticancer chemotherapeutic drug, which demonstrated beneficial effects in the CRPC treatment[168]. Blocking this interaction, using a CCL25 neutralization peptide antibody, impaired the antiapoptotic mechanisms, inducing PCa cells apoptosis. The combined treatment of CRPC mice xenografts with etoposide and CCL25 antibody significantly decreased the tumor burden, suggesting a beneficial effect of this peptide as an adjuvant therapy for CRPC[168]. The invasive ability of PCa cells was also impaired by disrupting PPIs using peptides. Indeed, the repression of Wiskott-Aldrich syndrome protein family member 3 (WASF3)/ Cytoplasmic FMR1-interacting protein 1 (CYFIP1) complex, using stapled peptides that target an α-helical interface, decreased PCa cells invasion i*n vitro*, indicating a beneficial role in inhibiting PCa metastasis[169].

## 5. Other PPIs with potential to be disrupted in PCa

Beyond the PPIs whose disruption has already been tested, others have been identified as promising therapeutic targets for PCa (**Table 3**). Besides interactions of AR with its coactivators, PPIs between AR and other proteins have been correlated with important roles in PCa progression[170–177]. As examples, the interactions of AR with signal transducer and activator of transcription 3 (STAT3) and peroxiredoxin-1 (PRDX1) were associated with increased AR transcriptional activity and PCa progression[170,171]. In addition, AR/ transcription factor SOX-9 (SOX9), AR/ N-acetyltransferase arrest-defect 1 protein (ARD1), and AR/ semenogelin I (SEMG1) interactions were associated with increased PCa cells proliferation[172–174]. Some protein phosphatases/ kinases have been considered AR coactivators[175,176]. In particular, serine/threonine-protein phosphatase PP1 (PP1), by interacting with AR, suppressed its ubiquitin-mediated proteasome degradation. The AR interaction with zipper interacting protein kinase (ZIPK) also increased AR-mediated transcriptional activity and ZIPK contributed to anti-apoptotic and proliferative functions of AR in PCa cells[177].

Additional interactions involving protein phosphatases/ kinases have also been considered as promising therapeutic targets for PCa, although their disruption has not yet been tested[178–182]. PP1 interacts with several proteins with important outcomes for PCa progression. The PP1/caveolin-1 (CAV1) interaction is associated with PP1 inhibition and was responsible for CAV1-mediated PCa cells survival[178]. In addition, the tyrosine-protein kinase Fer (FER) interaction with PP1 contributed to cell cycle progression of malignant PCa cells[179]. Finally, PP1 interacts with nuclear inhibitor of protein phosphatase 1 (NIPP1) and this interaction plays a key role to increase and direct the migration of PCa cells, thus contributing to metastasis[180]. A role in chemotherapeutic drug resistance was also evident for some PPIs involving protein kinases. In fact, both MST1/ HSP70 and protein kinase C (PKC)/ Ectonucleoside triphosphate diphosphohydrolase 5 (PCPH) interactions promoted resistance to cisplatin-induced PCa cells apoptosis[181,182]. The PKC/PCPH interaction was also associated with increased PCa invasiveness[182].

Other PPIs have also been considered potential therapeutic targets for PCa. The bradykinin receptor 1 (B1R)/ B2R, S100A9/ Toll-like receptor 4 (TLR4), Tripartite motif-containing protein 25 (TRIM25)/ GTPase-activating protein-binding protein 2 (G3BP2) and Runt-related

transcription factor 2 (RUNX2)/ SMAD interactions promoted PCa tumor growth[183–186]. Besides this effect, the interaction of RUNX2 with SMAD also controlled the PCa metastatic process[186]. Other PPIs, namely Annexin A2 (ANXA2)/ STAT6 and Cluster of differentiation 44 (CD44)/ Vascular cell adhesion molecule 1 (VCAM1) played an important role in PCa metastasis[187,188]. In particular, the interaction of CD44 with VCAM1 mediated the adhesion of PCa cells to vascular endothelial cells, a key initial step in metastatic process[188]. Lastly, the interaction between Prostate Leucine Zipper gene (PC1) and initiation factor 4E-binding protein 1 (4EBP1) conferred resistance to rapamycin treatment[189].

## 6. Concluding remarks and future perspectives

The disruption of PPIs has emerged as a promising approach for anticancer therapies. Nevertheless, the lack of cancer-specific analysis in PPIs databases has limited this approach. Besides, the process of developing a PPI inhibitor remains long and difficult. Interrupting AR-related PPIs seems to be a promising strategy since AR assumes a key role in PCa progression. Several small molecules were suggested for this purpose and beneficial results were observed either alone or in combination with other chemotherapeutic drugs. On the other hand, despite the potential of targeting Bcl-2-related PPIs to promote PCa cells apoptosis and despite the largest number of compounds developed to target them, most of the compounds lack selectivity and some were even considered PAINS. These findings have limited the development of compounds that effectively targets Bcl-2-associated PPIs, since most were associated with toxicity and off-target effects. Other small molecules that target kinases/phosphatases-related PPIs or other PPIs were also associated with promising results, although some are limited by target multiplicity or by affecting the Wnt pathway.

Peptides emerged more recently as a favourable strategy to target PPIs. Developed to circumvent the main limitations of small molecules, peptides have been associated with relatively few off-target effects. Most of the peptides developed to target PPIs were synthesized to mimic the structure of the PPI interface and often exhibit exquisite specificity and affinity to their targets. Nevertheless, most of them lack detailed target engagement data and more information is still required to confirm their potential.

Among the 28 small molecules and 14 peptides proposed for the treatment of PCa, some were associated with a role in CRPC treatment. In addition, we highlighted the potential role

of several compounds in the treatment of CRPC which interfere with signaling pathways dysregulated in this more aggressive cancer type. The discovery of new therapeutic options for CRPC remains a significant challenge of PCa treatment, but most of the compounds described to date were tested in cell lines, including PC-3 and LNCaP, that are not considered CRPC models. Similarly, some authors claim that the DU-145 cell line, used to evaluate some of the compounds described herein, is a useful model of CRPC. However, since this cell line does not express AR, the main driver of CRPC initiation and progression, any conclusions drawn from such studies are limited. Thus, ailanthone, UPF1069, SRC1-derived peptide and MST1 kinase were the only compounds tested in CRPC cell lines (22RV1 or C4-2) with promising results. Moreover, OGX-427 and metformin were successfully tested in phase I/II clinical trial for CRPC treatment and the phase I clinical trial of simvastatin is currently ongoing.

In conclusion, the targeting of PPIs as a PCa therapeutic option, either as a monotherapy or in combination with other therapeutic agents, remains a challenging, but promising approach and is a topic with many open doors. Maximizing the range of PPIs that can be targeted will be crucial to exploit the wide variety of intracellular molecular targets and potential drug targets of the future. Structure-based studies of PPI interfaces are also essential to improve the effectiveness of targeting strategies. Thus, more research and advancement are urgently required to develop improved assays to modulate PPIs and develop a new and effective therapeutic strategy for PCa and CRPC.

**Declarations of Interest:**

None

**Acknowledgments:**

We thank the Portuguese Foundation for Science and Technology(FCT), European Union, QREN, FEDER and COMPETE for funding iBiMED (UIDB/04501/2020, POCI-01-0145-FEDER-

007628 and UID/BIM/04501/2019) and an individual scholarship from BM (SFRH/BD/146032/2019).

**References**

[1] F. Bray, J. Ferlay, I. Soerjomataram, R.L. Siegel, L.A. Torre, A. Jemal, Global cancer statistics 2018: GLOBOCAN estimates of incidence and mortality worldwide for 36 cancers in 185 countries, CA. Cancer J. Clin. 68 (2018) 394–424. https://doi.org/10.3322/caac.21492.

[2] M.S. Litwin, H.J. Tan, The diagnosis and treatment of prostate cancer: A review, JAMA - J. Am. Med. Assoc. 317 (2017) 2532–2542. https://doi.org/10.1001/jama.2017.7248.

[3] E. Nevedomskaya, S.J. Baumgart, B. Haendler, Recent advances in prostate cancer treatment and drug discovery, Int. J. Mol. Sci. 19 (2018). https://doi.org/10.3390/ijms19051359.

[4] I. Coutinho, T.K. Day, W.D. Tilley, L.A. Selth, Androgen receptor signaling in castration-resistant prostate cancer: A lesson in persistence, Endocr. Relat. Cancer. 23 (2016) T179–T197. https://doi.org/10.1530/ERC-16-0422.

[5] K. Wang, H. Ruan, T. Xu, L. Liu, D. Liu, H. Yang, X. Zhang, K. Chen, Recent advances on the progressive mechanism and therapy in castration-resistant prostate cancer, (2018) 3167–3178.

[6] P. Toren, A. Zoubeidi, Targeting the PI3K / Akt pathway in prostate cancer : Challenges and opportunities ( Review ), (2014) 1793–1801. https://doi.org/10.3892/ijo.2014.2601.

[7] N.N. Yokoyama, S. Shao, B.H. Hoang, D. Mercola, X. Zi, Wnt signaling in castration-resistant prostate cancer : implications for therapy, 2 (2014) 27–44.

[8] L. Zhang, S. Yang, X. Chen, S. Stauffer, F. Yu, S.M. Lele, K. Fu, K. Datta, The Hippo Pathway Effector YAP Regulates Motility , Invasion , and Castration-Resistant Growth of Prostate Cancer Cells, 35 (2015) 1350–1362. https://doi.org/10.1128/MCB.00102-15.

[9] A.B. Blanchet, E. Carton, L. Golmard, O. Huillard, M. Vidal, A PK/PD study of Delta-4 abiraterone metabolite in metastatic castration-resistant prostate cancer patients, Pharmacol. Res. (2018). https://doi.org/10.1016/j.phrs.2018.08.016.

[10] A.M. Aghdam, A. Amiri, R. Salarinia, A. Masoudifar, F. Ghasemi, H. Mirzaei, MicroRNAs as Diagnostic, Prognostic, and Therapeutic Biomarkers in Prostate Cancer Arad, 29 (2019) 127–139.

[11] M. Dragomir, A.C.P. Mafra, S.M.G. Dias, C. Vasilescu, G.A. Calin, Using microRNA Networks to Understand Cancer, (2018) 1–18. https://doi.org/10.3390/ijms19071871.

[12] A.A. Ivanov, F.R. Khuri, H. Fu, Targeting protein–protein interactions as an anticancer strategy, Trends Pharmacol. Sci. 34 (2013) 393–400. https://doi.org/10.1016/j.tips.2013.04.007.

[13] K. Luck, A reference map of the human protein interactome reveals tissue-specific biological mechanisms, BioRxiv. (2018) 4–6.

[14] I.A. Kovács, K. Luck, K. Spirohn, Y. Wang, C. Pollis, S. Schlabach, W. Bian, D.K. Kim, N. Kishore, T. Hao, M.A. Calderwood, M. Vidal, A.L. Barabási, Network-based prediction of protein interactions, Nat. Commun. 10 (2019) 1–8. https://doi.org/10.1038/s41467-019-09177-y.

[15] A. Nohe, N.O. Petersen, Analyzing protein-protein interactions in cell membranes, BioEssays. 26 (2004) 196–203. https://doi.org/10.1002/bies.10380.

[16] J. Westermarck, J. Ivaska, G.L. Corthals, Identification of protein interactions involved in cellular signaling, Mol. Cell. Proteomics. 12 (2013) 1752–1763. https://doi.org/10.1074/mcp.R113.027771.

[17] L. Mabonga, A.P. Kappo, Peptidomimetics: A Synthetic Tool for Inhibiting Protein–Protein Interactions in Cancer, Int. J. Pept. Res. Ther. 26 (2020) 225–241. https://doi.org/10.1007/s10989-019-09831-5.

[18] C.J. Lord, C.J. Ryan, Robust genetic interactions in cancer are enriched in protein-protein interaction pairs A " discovery and validation " approach to the analysis of loss-of-function, (2019) 6–11.

[19] Z. Li, A.A. Ivanov, R. Su, V. Gonzalez-Pecchi, Q. Qi, S. Liu, P. Webber, E. McMillan, L. Rusnak, C. Pham, X. Chen, X. Mo, B. Revennaugh, W. Zhou, A. Marcus, S. Harati, X. Chen, M.A. Johns, M.A. White, C. Moreno, L.A.D. Cooper, Y. Du, F.R. Khuri, H. Fu, The OncoPPi network of cancer-focused protein-protein interactions to inform biological insights and therapeutic strategies, Nat. Commun. 8 (2017) 1–13. https://doi.org/10.1038/ncomms14356.

[20] A.A. Ivanov, B. Revennaugh, L. Rusnak, V. Gonzalez-Pecchi, X. Mo, M.A. Johns, Y. Du, L.A.D. Cooper, C.S. Moreno, F.R. Khuri, H. Fu, The OncoPPi Portal: An integrative resource to explore and prioritize protein-protein interactions for cancer target discovery, Bioinformatics. 34 (2018) 1183–1191. https://doi.org/10.1093/bioinformatics/btx743.

[21] G. Gulfidan, B. Turanli, H. Beklen, R. Sinha, K.Y. Arga, Pan-cancer mapping of differential protein-protein interactions, Sci. Rep. 10 (2020) 1–12. https://doi.org/10.1038/s41598-020-60127-x.

[22] H. Billur Engin, E. Guney, O. Keskin, B. Oliva, A. Gursoy, Integrating structure to protein-protein interaction networks that drive metastasis to brain and lung in breast cancer, PLoS One. 8 (2013) 1–15. https://doi.org/10.1371/journal.pone.0081035.

[23] L.O. Sillerud, R.S. Larson, Design and Structure of Peptide and Peptidomimetic Antagonists of Protein- Protein Interaction, (2005) 151–169.

[24] A. Goncearenco, M. Li, F.L. Simonetti, B.A. Shoemaker, A.R. Panchenko, Exploring Protein-Protein Interactions as Drug Targets for Anti-cancer Therapy with In Silico Workflows, in: I.M. Lazar, M. Kontoyianni, A.C. Lazar (Eds.), Proteomics Drug Discov., Springer New York, New York, NY, 2017: pp. 221–236. https://doi.org/10.1007/978-1-4939-7201-2_15.

[25] X. Cao, J.L. Yap, M.K. Newell-Rogers, C. Peddaboina, W. Jiang, H.T. Papaconstantinou, D. Jupitor, A. Rai, K.Y. Jung, R.P. Tubin, W. Yu, K. Vanommeslaeghe, P.T. Wilder, A.D. MacKerell, S. Fletcher, R.W. Smythe, The novel BH3 α-helix mimetic JY-1-106 induces apoptosis in a subset of cancer cells (lung cancer, colon cancer and mesothelioma) by disrupting Bcl-xL and Mcl-1 protein-protein interactions with Bak., Mol. Cancer. 12

(2013) 1–16. https://doi.org/10.1186/1476-4598-12-42.

[26] T. Zhang, A. Hamza, X. Cao, B. Wang, S. Yu, C.G. Zhan, D. Sun, A novel Hsp90 inhibitor to disrupt Hsp90/Cdc37 complex against pancreatic cancer cells, Mol. Cancer Ther. 7 (2008) 162–170. https://doi.org/10.1158/1535-7163.MCT-07-0484.

[27] J. Shi, Y. Wang, L. Zeng, Y. Wu, J. Deng, Q. Zhang, Y. Lin, J. Li, T. Kang, M. Tao, E. Rusinova, G. Zhang, C. Wang, H. Zhu, J. Yao, Y.X. Zeng, B.M. Evers, M.M. Zhou, B.P. Zhou, Disrupting the Interaction of BRD4 with Diacetylated Twist Suppresses Tumorigenesis in Basal-like Breast Cancer, Cancer Cell. 25 (2014) 210–225. https://doi.org/10.1016/j.ccr.2014.01.028.

[28] C. Van Poznak, A.D. Seidman, M.M. Reidenberg, M.M. Moasser, N. Sklarin, K. Van Zee, P. Borgen, M. Gollub, D. Bacotti, T.J. Yao, R. Bloch, M. Ligueros, M. Sonenberg, L. Norton, C. Hudis, Oral gossypol in the treatment of patients with refractory metastatic breast cancer: A phase I/II clinical trial, Breast Cancer Res. Treat. 66 (2001) 239–248. https://doi.org/10.1023/A:1010686204736.

[29] G. Liu, W.K. Kelly, G. Wilding, L. Leopold, K. Brill, B. Somer, An Open-Label, Multicenter, Phase I/II Study of Single-Agent AT-101 in Men with Castrate-Resistant Prostate Cancer, Clin. Cancer Res. 15 (2009) 3172–3176. https://doi.org/10.1158/1078-0432.CCR-08-2985.

[30] J.J. Hwang, J. Kuruvilla, D. Mendelson, M.J. Pishvaian, J.F. Deeken, L.L. Siu, M.S. Berger, J. Viallet, J.L. Marshall, Phase I Dose Finding Studies of Obatoclax (GX15-070), a Small Molecule Pan-BCL-2 Family Antagonist, in Patients with Advanced Solid Tumors or Lymphoma, Clin. Cancer Res. 16 (2010) 4038–4045. https://doi.org/10.1158/1078-0432.CCR-10-0822.

[31] A.W. Roberts, J.F. Seymour, J.R. Brown, W.G. Wierda, T.J. Kipps, S.L. Khaw, D.A. Carney, S.Z. He, D.C.S. Huang, H. Xiong, Y. Cui, T.A. Busman, E.M. McKeegan, A.P. Krivoshik, S.H. Enschede, R. Humerickhouse, Substantial susceptibility of chronic lymphocytic leukemia to BCL2 inhibition: Results of a phase I study of navitoclax in patients with relapsed or refractory disease, J. Clin. Oncol. 30 (2012) 488–496. https://doi.org/10.1200/JCO.2011.34.7898.

[32] L. Gandhi, D.R. Camidge, M.R. De Oliveira, P. Bonomi, D. Gandara, D. Khaira, C.L. Hann, E.M. McKeegan, E. Litvinovich, P.M. Hemken, C. Dive, S.H. Enschede, C. Nolan, Y.L. Chiu, T. Busman, H. Xiong, A.P. Krivoshik, R. Humerickhouse, G.I. Shapiro, C.M. Rudin, Phase I study of navitoclax (ABT-263), a novel bcl-2 family inhibitor, in patients with small-cell lung cancer and other solid tumors, J. Clin. Oncol. 29 (2011) 909–916. https://doi.org/10.1200/JCO.2010.31.6208.

[33] C.M. Rudin, C.L. Hann, E.B. Garon, M. Ribeiro de Oliveira, P.D. Bonomi, D.R. Camidge, Q. Chu, G. Giaccone, D. Khaira, S.S. Ramalingam, M.R. Ranson, C. Dive, E.M. McKeegan, B.J. Chyla, B.L. Dowell, A. Chakravartty, C.E. Nolan, N. Rudersdorf, T.A. Busman, M.H. Mabry, A.P. Krivoshik, R.A. Humerickhouse, G.I. Shapiro, L. Gandhi, Phase II Study of Single-Agent Navitoclax (ABT-263) and Biomarker Correlates in Patients with Relapsed Small Cell Lung Cancer, Clin. Cancer Res. 18 (2012) 3163–3169. https://doi.org/10.1158/1078-0432.CCR-11-3090.

[34] M. Konopleva, D.A. Pollyea, J. Potluri, B.J. Chyla, T. Busman, E. McKeegan, A. Salem, M. Zhu, J.L. Ricker, W. Blum, C.D. DiNardo, M. Dunbar, R. Kirby, N. Falotico, J.D. Leverson, R.A. Humerickhouse, M. Mabry, R.M. Stone, H.M. Kantarjian, A.G. Letai, A Phase 2 Study of ABT-199 (GDC-0199) in Patients with Acute Myelogenous Leukemia (AML), Blood. 124 (2014) 118–118. https://doi.org/10.1182/blood.V124.21.118.118.

[35] A. Patnaik, A. Tolcher, M. Beeram, J. Nemunaitis, G.J. Weiss, K. Bhalla, M. Agrawal, G. Nichols, S. Middleton, A. Beryozkina, N. Sarapa, R. Peck, J. Zhi, Clinical pharmacology characterization of RG7112, an MDM2 antagonist, in patients with advanced solid tumors, Cancer Chemother. Pharmacol. 76 (2015) 587–595. https://doi.org/10.1007/s00280-015-2830-8.

[36] M. Andreeff, K.R. Kelly, K. Yee, S. Assouline, R. Strair, L. Popplewell, D. Bowen, G. Martinelli, M.W. Drummond, P. Vyas, M. Kirschbaum, S.P. Iyer, V. Ruvolo, G.M.N. Gonzalez, X. Huang, G. Chen, B. Graves, S. Blotner, P. Bridge, L. Jukofsky, S. Middleton, M. Reckner, R. Rueger, J. Zhi, G. Nichols, K. Kojima, Results of the phase i trial of RG7112, a small-molecule MDM2 antagonist in leukemia, Clin. Cancer Res. 22 (2016) 868–876. https://doi.org/10.1158/1078-0432.CCR-15-0481.

[37] K. Yee, G. Martinelli, N. Vey, M.J. Dickinson, K. Seiter, S. Assouline, M. Drummond, S.-

S. Yoon, M. Kasner, J.-H. Lee, K.R. Kelly, S. Blotner, B. Higgins, S. Middleton, G. Nichols, G. Chen, H. Zhong, W.E. Pierceall, J. Zhi, L.-C. Chen, Phase 1/1b Study of RG7388, a Potent MDM2 Antagonist, in Acute Myelogenous Leukemia (AML) Patients (Pts), Blood. 124 (2014) 116–116. https://doi.org/10.1182/blood.V124.21.116.116.

[38] T. Bauer, D. Hong, N. Somaiah, C. Cai, S. Song, P. Kumar, R. Gajee, M. Rosen, J. Kochan, S. Chen, D. Hyman, T. Masters, F. Meric-Bernstam, A. Tse, P. LoRusso, A. Weise, M. Gounder, Abstract B27: A phase I dose escalation study of the MDM2 inhibitor DS-3032b in patients with advanced solid tumors and lymphomas, in: Clin. Trials, American Association for Cancer Research, 2015: pp. B27–B27. https://doi.org/10.1158/1535-7163.TARG-15-B27.

[39] A.J. Wagner, U. Banerji, A. Mahipal, N. Somaiah, H.A. Hirsch, C. Fancourt, A. Levonas, R. Lam, A. Meister, R.K. Kemp, C. Knox, S. Rose, D.S. Hong, A phase I trial of the human double minute 2 (HDM2) inhibitor MK-8242 in patients (pts) with advanced solid tumors., J. Clin. Oncol. 33 (2015) 10564–10564. https://doi.org/10.1200/jco.2015.33.15_suppl.10564.

[40] F. Ravandi, I. Gojo, M.M. Patnaik, M.D. Minden, H.M. Kantarjian, A. Levonas, C. Fancourt, R. Lam, A. Meister, M.E. Jones, R.K. Kemp, C. Knox, S. Rose, P. Shah Patel, R. Tibes, A phase I trial of the human double minute 2 inhibitor (MK-8242) in patients with refractory/recurrent acute myelogenous leukemia (AML)., J. Clin. Oncol. 33 (2015) 7070–7070. https://doi.org/10.1200/jco.2015.33.15_suppl.7070.

[41] H.I. Hurwitz, D.C. Smith, H.C. Pitot, J.M. Brill, R. Chugh, E. Routits, J. Rubin, J. Strickler, G. Vuagniaux, J.M. Sorensen, C. Zanna, Safety, pharmacokinetics, and pharmacodynamic properties of oral DEBIO1143 (AT-406) in patients with advanced cancer: Results of a first-in-man study, Cancer Chemother. Pharmacol. 75 (2015) 851–859. https://doi.org/10.1007/s00280-015-2709-8.

[42] J.A. Flygare, M. Beresini, N. Budha, H. Chan, I.T. Chan, S. Cheeti, F. Cohen, K. Deshayes, K. Doerner, S.G. Eckhardt, L.O. Elliott, B. Feng, M.C. Franklin, S.F. Reisner, L. Gazzard, J. Halladay, S.G. Hymowitz, H. La, P. LoRusso, B. Maurer, L. Murray, E. Plise, C. Quan, J. Stephan, S.G. Young, J. Tom, V. Tsui, J. Um, E. Varfolomeev, D. Vucic, A.J. Wagner, H.J.A. Wallweber, L. Wang, J. Ware, Z. Wen, H. Wong, J.M. Wong, M. Wong, S. Wong, R. Yu,

K. Zobel, W.J. Fairbrother, Discovery of a Potent Small-Molecule Antagonist of Inhibitor of Apoptosis (IAP) Proteins and Clinical Candidate for the Treatment of Cancer (GDC-0152), J. Med. Chem. 55 (2012) 4101–4113. https://doi.org/10.1021/jm300060k.

[43] A.W. Tolcher, K.P. Papadopoulos, A. Patnaik, W.J. Fairbrother, H. Wong, N.R. Budha, W.C. Darbonne, F. V. Peale, M.J. Mamounas, S. Royer-Joo, R. Yu, C.C. Portera, J.C. Bendell, H.A. Burris, J.R. Infante, Phase I study of safety and pharmacokinetics (PK) of GDC-0917, an antagonist of inhibitor of apoptosis (IAP) proteins in patients (Pts) with refractory solid tumors or lymphoma., J. Clin. Oncol. 31 (2013) 2503–2503. https://doi.org/10.1200/jco.2013.31.15_suppl.2503.

[44] B.I. Sikic, S.G. Eckhardt, G. Gallant, H.A. Burris, D.R. Camidge, A.D. Colevas, S.F. Jones, W.A. Messersmith, H.A. Wakelee, H. Li, P.G. Kaminker, S. Morris, J.R. Infante, Safety, pharmacokinetics (PK), and pharmacodynamics (PD) of HGS1029, an inhibitor of apoptosis protein (IAP) inhibitor, in patients (Pts) with advanced solid tumors: Results of a phase I study., J. Clin. Oncol. 29 (2011) 3008–3008. https://doi.org/10.1200/jco.2011.29.15_suppl.3008.

[45] J. Tabernero, L. Dirix, P. Schöfski, A. Cervantes, J.A. Lopez-Martin, J. Capdevila, L. Van Beijsterveldt, S. Platero, B. Hall, Z. Yuan, R. Knoblauch, S.H. Zhuang, A phase I first-in-human pharmacokinetic and pharmacodynamic study of serdemetan in patients with advanced solid tumors, Clin. Cancer Res. 17 (2011) 6313–6321. https://doi.org/10.1158/1078-0432.CCR-11-1101.

[46] L. Jin, W. Wang, G. Fang, Targeting Protein-Protein Interaction by Small Molecules, Annu Rev Pharmacol Toxicol. (2014). https://doi.org/10.1146/annurev-pharmtox-011613-140028.

[47] D.E. Scott, A.R. Bayly, C. Abell, J. Skidmore, Small molecules, big targets: Drug discovery faces the protein-protein interaction challenge, Nat. Rev. Drug Discov. 15 (2016) 533–550. https://doi.org/10.1038/nrd.2016.29.

[48] C. Dai, H. Heemers, N. Sharifi, Androgen signaling in prostate cancer, Cold Spring Harb. Perspect. Med. 7 (2017). https://doi.org/10.1101/cshperspect.a030452.

[49] L. Galbraith, H.Y. Leung, I. Ahmad, Lipid Pathway Deregulation in Advanced Prostate

Cancer, Pharmacol. Res. (2018). https://doi.org/10.1016/j.phrs.2018.02.022.

[50] S. Yang, M. Jiang, M.M. Grabowska, J. Li, Z.M. Connelly, J. Zhang, S.W. Hayward, J.M. Cates, G. Han, X. Yu, Androgen receptor differentially regulates the proliferation of prostatic epithelial cells *in vitro* and *in vivo*, Oncotarget. 7 (2016). https://doi.org/10.18632/oncotarget.11879.

[51] E.D. Crawford, P.F. Schellhammer, D.G. McLeod, J.W. Moul, C.S. Higano, N. Shore, L. Denis, P. Iversen, M.A. Eisenberger, F. Labrie, Androgen Receptor Targeted Treatments of Prostate Cancer: 35 Years of Progress with Antiandrogens, J. Urol. 200 (2018) 956–966. https://doi.org/10.1016/j.juro.2018.04.083.

[52] C.A. Heinlein, C. Chang, Androgen receptor (AR) coregulators: An overview, Endocr. Rev. 23 (2002) 175–200. https://doi.org/10.1210/edrv.23.2.0460.

[53] K. Nikhil, S. Sharan, A. Chakraborty, P. Roy, Pterostilbene-isothiocyanate conjugate suppresses growth of prostate cancer cells irrespective of androgen receptor status, PLoS One. 9 (2014). https://doi.org/10.1371/journal.pone.0093335.

[54] E. Lee, A. Madar, G. David, M.J. Garabedian, R. Das Gupta, S.K. Logan, Inhibition of androgen receptor and β-catenin activity in prostate cancer, Proc. Natl. Acad. Sci. U. S. A. 110 (2013) 15710–15715. https://doi.org/10.1073/pnas.1218168110.

[55] M. Kahn, Can we safely target the WNT pathway? Michael, 13 (2015) 513–532. https://doi.org/10.1038/nrd4233.Can.

[56] M.B. Stope, T. Schubert, D. Staar, C. Rönnau, A. Streitbörger, N. Kroeger, C. Kubisch, U. Zimmermann, R. Walther, M. Burchardt, Effect of the heat shock protein HSP27 on androgen receptor expression and function in prostate cancer cells, World J. Urol. 30 (2012) 327–331. https://doi.org/10.1007/s00345-012-0843-z.

[57] Y. He, S. Peng, J. Wang, H. Chen, X. Cong, A. Chen, M. Hu, M. Qin, H. Wu, S. Gao, L. Wang, X. Wang, Z. Yi, M. Liu, Ailanthone targets p23 to overcome MDV3100 resistance in castration-resistant prostate cancer, Nat. Commun. 7 (2016) 1–14. https://doi.org/10.1038/ncomms13122.

[58] A. Zoubeidi, A. Zardan, E. Beraldi, L. Fazli, R. Sowery, P. Rennie, C. Nelson, M.E. Gleave,

Cooperative interactions between androgen receptor (AR) and heat-shock protein 27 facilitate AR transcriptional activity, Cancer Res. 67 (2007) 10455–10465. https://doi.org/10.1158/0008-5472.CAN-07-2057.

[59] K.N. Chi, E.Y. Yu, C. Jacobs, J. Bazov, C. Kollmannsberger, C.S. Higano, S.D. Mukherjee, M.E. Gleave, P.S. Stewart, S.J. Hotte, A phase I dose-escalation study of apatorsen (OGX-427), an antisense inhibitor targeting heat shock protein 27 (Hsp27), in patients with castration-resistant prostate cancer and other advanced cancers, Ann. Oncol. 27 (2016) 1116–1122. https://doi.org/10.1093/annonc/mdw068.

[60] F.N. Hsu, M.C. Chen, K.C. Lin, Y.T. Peng, P.C. Li, E. Lin, M.C. Chiang, J.T. Hsieh, H. Lin, Cyclin-dependent kinase 5 modulates STAT3 and androgen receptor activation through phosphorylation of ser727 on STAT3 in prostate cancer cells, Am. J. Physiol. - Endocrinol. Metab. 305 (2013). https://doi.org/10.1152/ajpendo.00615.2012.

[61] F.N. Hsu, M.C. Chen, M.C. Chiang, E. Lin, Y.T. Lee, P.H. Huang, G.S. Lee, H. Lin, Regulation of androgen receptor and prostate cancer growth by cyclin-dependent kinase 5, J. Biol. Chem. 286 (2011) 33141–33149. https://doi.org/10.1074/jbc.M111.252080.

[62] G. Wang, J. Wang, S. Marianne, Crosstalk between the Androgen Receptor and β-Catenin in Castrate Resistant Prostate Cancer, 68 (2009) 9918–9927. https://doi.org/10.1158/0008-5472.CAN-08-1718.

[63] B.C. Koehler, A.L. Scherr, S. Lorenz, T. Urbanik, N. Kautz, C. Elssner, S. Welte, J.L. Bermejo, D. Jäger, H. Schulze-Bergkamen, Beyond Cell Death - Antiapoptotic Bcl-2 Proteins Regulate Migration and Invasion of Colorectal Cancer Cells In Vitro, PLoS One. 8 (2013). https://doi.org/10.1371/journal.pone.0076446.

[64] J. Choi, K. Choi, E.N. Benveniste, Y.S. Hong, J.H. Lee, J. Kim, K. Park, Bcl-2 promotes invasion and lung metastasis by inducing matrix metalloproteinase-2, Cancer Res. 65 (2005) 5554–5560. https://doi.org/10.1158/0008-5472.CAN-04-4570.

[65] D. Trisciuoglio, M. Desideri, L. Ciuffreda, M. Mottolese, D. Ribatti, A. Vacca, M. Del Rosso, L. Marcocci, G. Zupi, D. Del Bufalo, Bcl-2 overexpression in melanoma cells increases tumor progression- associated properties and in vivo tumor growth, J. Cell.

Physiol. 205 (2005) 414–421. https://doi.org/10.1002/jcp.20413.

[66] A.J. Raffo, H. Perlman, M.W. Chen, J.S. Streitman, R. Buttyan, M.L. Day, Overexpression of bcl-2 Protects Prostate Cancer Cells from Apoptosis in Vitro and Confers Resistance to Androgen Depletion in Vivo, Cancer Res. 55 (1995) 4438–4445.

[67] G. Zinzalla, D.E. Thurston, Targeting protein-protein interactions for therapeutic intervention: A challenge for the future, Future Med. Chem. 1 (2009) 65–93. https://doi.org/10.4155/fmc.09.12.

[68] Y. Lin, Y. Lin, J. Fukuchi, R.A. Hiipakka, J.M. Kokontis, J. Xiang, Up-regulation of Bcl-2 is required for the progression of prostate cancer cells from an androgen-dependent to an androgen-independent growth stage, (2007) 531–536. https://doi.org/10.1038/cr.2007.12.

[69] D.H. Lee, M. Szczepanski, Y.J. Lee, Role of Bax in quercetin-induced apoptosis in human prostate cancer cells, Biochem. Pharmacol. 75 (2008) 2345–2355. https://doi.org/10.1016/j.bcp.2008.03.013.

[70] X. Lu, F. Yang, D. Chen, Q. Zhao, D. Chen, H. Ping, N. Xing, Quercetin reverses docetaxel resistance in prostate cancer via androgen receptor and PI3K / Akt signaling pathways, 16 (2020). https://doi.org/10.7150/ijbs.41686.

[71] Y. Meng, W. Tang, Y. Dai, X. Wu, M. Liu, Q. Ji, M. Ji, K. Pienta, T. Lawrence, L. Xu, Natural BH3 mimetic (-)-gossypol chemosensitizes human prostate cancer via Bcl-xL inhibition accompanied by increase of Puma and Noxa, Mol. Cancer Ther. 7 (2008) 2192–2202. https://doi.org/10.1158/1535-7163.MCT-08-0333.

[72] L. Xu, D. Yang, S. Wang, W. Tang, M. Liu, M. Davis, J. Chen, J.M. Rae, T. Lawrence, M.E. Lippman, ( À ) -Gossypol enhances response to radiation therapy and results in tumor regression of human prostate cancer, 197 (2005) 197–206.

[73] J. Glaser, U. Holzgrabe, Focus on PAINS: false friends in the quest for selective anti-protozoal lead structures from, Medchemcomm. (2016). https://doi.org/10.1039/C5MD00481K.

[74] M.K. Matlock, T.B. Hughes, J.L. Dahlin, S.J. Swamidass, Modeling Small-Molecule

Reactivity Identi fi es Promiscuous Bioactive Compounds, (2018). https://doi.org/10.1021/acs.jcim.8b00104.

[75] J. Glaser, M. Schultheis, S. Hazra, B. Hazra, H. Moll, U. Schurigt, U. Holzgrabe, Antileishmanial Lead Structures from Nature: Analysis of Structure-Activity Relationships of a Compound Library Derived from Caffeic Acid Bornyl Ester, (2014) 1394–1410. https://doi.org/10.3390/molecules19021394.

[76] C.W. Shiau, J.W. Huang, D.S. Wang, J.R. Weng, C.C. Yang, C.H. Lin, C. Li, C.S. Chen, α-tocopheryl succinate induces apoptosis in prostate cancer cells in part through inhibition of Bcl-xL/Bcl-2 function, J. Biol. Chem. 281 (2006) 11819–11825. https://doi.org/10.1074/jbc.M511015200.

[77] M.A. Savitskaya, G.E. Onischenko, α Tocopheryl Succinate Affects Malignant Cell Viability , Proliferation , and Differentiation, 81 (2016) 806–818.

[78] M. Bruncko, T.K. Oost, B.A. Belli, H. Ding, M.K. Joseph, A. Kunzer, D. Martineau, W.J. McClellan, M. Mitten, S.C. Ng, P.M. Nimmer, T. Oltersdorf, C.M. Park, A.M. Petros, A.R. Shoemaker, X. Song, X. Wang, M.D. Wendt, H. Zhang, S.W. Fesik, S.H. Rosenberg, S.W. Elmore, Studies leading to potent, dual inhibitors of Bcl-2 and Bcl-xL, J. Med. Chem. 50 (2007) 641–662. https://doi.org/10.1021/jm061152t.

[79] R. Parrondo, A. de las Pozas, T. Reiner, C. Perez-Stable, ABT-737, a small molecule Bcl-2/Bcl-xL antagonist, increases antimitoticmediated apoptosis in human prostate cancer cells, PeerJ. 2013 (2013) 1–22. https://doi.org/10.7717/peerj.144.

[80] T. Noll, S. Schultze, S. Irina, K. Marta, M. Philipp, Synergistic cytotoxicity of a prostate cancer-specific immunotoxin in combination with the BH3 mimetic ABT-737, Cancer Immunol. Immunother. 67 (2018) 413–422. https://doi.org/10.1007/s00262-017-2097-5.

[81] C. Tse, A.R. Shoemaker, J. Adickes, M.G. Anderson, J. Chen, S. Jin, E.F. Johnson, K.C. Marsh, M.J. Mitten, P. Nimmer, L. Roberts, S.K. Tahir, Y. Xiao, X. Yang, H. Zhang, S. Fesik, S.H. Rosenberg, S.W. Elmore, ABT-263 : A Potent and Orally Bioavailable Bcl-2 Family Inhibitor, (2008) 3421–3429. https://doi.org/10.1158/0008-5472.CAN-07-5836.

[82] D. Xiao, S. Choi, D.E. Johnson, V.G. Vogel, C.S. Johnson, D.L. Trump, Y.J. Lee, S. V. Singh,

Diallyl trisulfide-induced apoptosis in human prostate cancer cells involves c-Jun N-terminal kinase and extracellular-signal regulated kinase-mediated phosphorylation of Bcl-2, Oncogene. 23 (2004) 5594–5606. https://doi.org/10.1038/sj.onc.1207747.

[83] S. V Singh, A.A. Powolny, S.D. Stan, D. Xiao, J.A. Arlotti, R. Warin, E. Hahm, S.W. Marynowski, A. Bommareddy, M. Potter, R. Dhir, Garlic Constituent Diallyl Trisulfide Prevents Development of Poorly-Differentiated Prostate Cancer and Pulmonary Metastasis, 68 (2009) 9503–9511. https://doi.org/10.1158/0008-5472.CAN-08-1677.Garlic.

[84] S.D. Stan, S. V Singh, Transcriptional Repression and Inhibition of Nuclear Translocation of Androgen Receptor by Diallyl Trisulfide in Human Prostate Cancer Cells, 15 (2009) 4895–4904. https://doi.org/10.1158/1078-0432.CCR-09-0512.

[85] A. Borkowska, N. Knap, J. Antosiewicz, Diallyl Trisulfide Is More Cytotoxic to Prostate Cancer Cells PC-3 than to Noncancerous Epithelial Cell Line PNT1A : A Possible Role of p66Shc signaling Axis Diallyl Trisulfide Is More Cytotoxic to Prostate Cancer Cells PC-3 than to Noncancerous Epithelial Cell Line PNT1A : A Possible Role of p66Shc signaling Axis, (n.d.) 37–41. https://doi.org/10.1080/01635581.2013.789115.

[86] J. Lian, X. Wu, F. He, D. Karnak, W. Tang, Y. Meng, D. Xiang, M. Ji, T.S. Lawrence, L. Xu, A natural BH3 mimetic induces autophagy in apoptosis-resistant prostate cancer via modulating Bcl-2-Beclin1 interaction at endoplasmic reticulum, Cell Death Differ. 18 (2011) 60–71. https://doi.org/10.1038/cdd.2010.74.

[87] B. Rah, R. ur Rasool, D. Nayak, S.K. Yousuf, D. Mukherjee, L.D. Kumar, A. Goswami, PAWR-mediated suppression of BCL2 promotes switching of 3-azido withaferin A (3-AWA)-induced autophagy to apoptosis in prostate cancer cells, Autophagy. 11 (2015) 314–331. https://doi.org/10.1080/15548627.2015.1017182.

[88] J. Decuypere, J.B. Parys, G. Bultynck, Regulation of the Autophagic Bcl-2/Beclin 1 Interaction, (2012) 284–312. https://doi.org/10.3390/cells1030284.

[89] B. Rah, H. Amin, K. Yousuf, S. Khan, G. Jamwal, D. Mukherjee, A Novel MMP-2 Inhibitor 3-azidowithaferin A ( 3- azidoWA ) Abrogates Cancer Cell Invasion and Angiogenesis by Modulating Extracellular Par-4, 7 (2012).

https://doi.org/10.1371/journal.pone.0044039.

[90] E. Seo, S. Kim, E. Jho, Induction of cancer cell-specific death via MMP2 promoter-dependent Bax expression, (n.d.) 217–222.

[91] S. Shukla, P. Fu, S. Gupta, Apigenin induces apoptosis by targeting inhibitor of apoptosis proteins and Ku70-Bax interaction in prostate cancer, Apoptosis. 19 (2014) 883–894. https://doi.org/10.1007/s10495-014-0971-6.

[92] P. Kaur, S. Shukla, S. Gupta, Plant flavonoid apigenin inactivates Akt to trigger apoptosis in human prostate cancer: An in vitro and in vivo study, Carcinogenesis. 29 (2008) 2210–2217. https://doi.org/10.1093/carcin/bgn201.

[93] D. Xiao, S. V. Singh, Diallyl trisulfide, a constituent of processed garlic, inactivates Akt to trigger mitochondrial translocation of BAD and caspase-mediated apoptosis in human prostate cancer cells, Carcinogenesis. 27 (2006) 533–540. https://doi.org/10.1093/carcin/bgi228.

[94] M. Sawada, W. Sun, P. Hayes, K. Leskov, D.A. Boothman, S. Matsuyama, Ku70 suppresses the apoptotic translocation of bax to mitochondria, Nat. Cell Biol. 5 (2003) 320–329. https://doi.org/10.1038/ncb950.

[95] M. Chien, Y. Lin, Y. Wen, Y. Yang, M. Hsiao, J. Chang, Targeting the SPOCK1-snail / slug axis- mediated epithelial-to-mesenchymal transition by apigenin contributes to repression of prostate cancer metastasis, 3 (2019) 1–17.

[96] J.H. Bushweller, Targeting transcription factors in cancer — from undruggable to reality, Nat. Rev. Cancer. 19 (2019) 611–624. https://doi.org/10.1038/s41568-019-0196-7.

[97] K.A. Fitzgerald, J.C. Evans, J. McCarthy, J. Guo, M. Prencipe, M. Kearney, W.R. Watson, C.M. O'Driscoll, The role of transcription factors in prostate cancer and potential for future RNA interference therapy, Expert Opin. Ther. Targets. 18 (2014) 633–649. https://doi.org/10.1517/14728222.2014.896904.

[98] S. Patiar, A.L. Harris, Role of hypoxia-inducible factor-1α as a cancer therapy target, Endocr. Relat. Cancer. 13 (2006) 61–75. https://doi.org/10.1677/erc.1.01290.

[99] G.L. Semenza, Defining the role of hypoxia-inducible factor 1 in cancer biology and therapeutics, Oncogene. 29 (2010) 625–634. https://doi.org/10.1038/onc.2009.441.

[100] J.C. Jun, A. Rathore, H. Younas, D. Gilkes, V.Y. Polotsky, Hypoxia-Inducible Factors and Cancer, Curr. Sleep Med. Reports. 3 (2017) 1–10. https://doi.org/10.1007/s40675-017-0062-7.

[101] C. Zou, S. Yu, Z. Xu, D. Wu, C.F. Ng, X. Yao, D.T. Yew, J.M. Vanacker, F.L. Chan, ERRα augments HIF-1 signalling by directly interacting with HIF-1 α in normoxic and hypoxic prostate cancer cells, J. Pathol. 233 (2014) 61–73. https://doi.org/10.1002/path.4329.

[102] K.M. Reece, E.D. Richardson, K.M. Cook, T.J. Campbell, S.T. Pisle, A.J. Holly, D.J. Venzon, D.J. Liewehr, C.H. Chau, D.K. Price, W.D. Figg, Epidithiodiketopiperazines (ETPs) exhibit in vitro antiangiogenic and in vivo antitumor activity by disrupting the HIF-1α/p300 complex in a preclinical model of prostate cancer, Mol. Cancer. 13 (2014) 1–12. https://doi.org/10.1186/1476-4598-13-91.

[103] D. Schlam, J. Canton, M. Carreño, H. Kopinski, S.A. Freeman, S. Grinstein, Gliotoxin Suppresses Macrophage Immune Function by Subverting, 7 (2016) 1–15. https://doi.org/10.1128/mBio.02242-15.Editor.

[104] J.A. Schneider, S.K. Logan, Revisiting the role of Wnt/β-catenin signaling in prostate cancer, Mol. Cell. Endocrinol. (2017). https://doi.org/10.1016/j.mce.2017.02.008.

[105] X. He, W. Zhang, C. Yan, F. Nie, C. Li, X. Liu, C. Fei, S. Li, Chemical biology reveals CARF as a positive regulator of canonical Wnt signaling by promoting TCF / β -catenin transcriptional activity, Nat. Publ. Gr. 3 (2017) 1–15. https://doi.org/10.1038/celldisc.2017.3.

[106] S. Gu, P. Hou, K. Liu, X. Niu, B. Wei, F. Mao, Z. Xu, ZNF433 positively regulates the beta-catenin/ tcf pathway in prostate cancer and enhances the tumorigenicity of cancer cells, Onco. Targets. Ther. 12 (2019) 1031–1039. https://doi.org/10.2147/OTT.S178150.

[107] R. Rebello, R. Pearson, R. Hannan, L. Furic, Therapeutic Approaches Targeting MYC-Driven Prostate Cancer, (2017). https://doi.org/10.3390/genes8020071.

[108] A. Castell, Q. Yan, K. Fawkner, P. Hydbring, F. Zhang, V. Verschut, M. Franco, S.M. Zakaria, W. Bazzar, J. Goodwin, G. Zinzalla, L.G. Larsson, A selective high affinity MYC-binding compound inhibits MYC:MAX interaction and MYC-dependent tumor cell proliferation, Sci. Rep. 8 (2018) 1–17. https://doi.org/10.1038/s41598-018-28107-4.

[109] J. Guo, R.A. Parise, E. Joseph, M.J. Egorin, J.S. Lazo, E. V. Prochownik, J.L. Eiseman, Efficacy, pharmacokinetics, tisssue distribution, and metabolism of the Myc-Max disruptor, 10058-F4 [Z,E]-5-[4-ethylbenzylidine]-2-thioxothiazolidin-4- one, in mice, Cancer Chemother. Pharmacol. 63 (2009) 615–625. https://doi.org/10.1007/s00280-008-0774-y.

[110] S. Fletcher, E. Prochownik, Small-Molecule Inhibitors of the Myc Oncoprotein, 2016. https://doi.org/10.1016/j.bbagrm.2014.03.005.Small-Molecule.

[111] S. Bai, S. Cao, L. Jin, M. Kobelski, B. Schouest, X. Wang, N. Ungerleider, M. Baddoo, W. Zhang, E. Corey, R.L. Vessella, X. Dong, K. Zhang, X. Yu, E.K. Flemington, Y. Dong, A positive role of c-Myc in regulating androgen receptor and its splice variants in prostate cancer, Oncogene. (n.d.). https://doi.org/10.1038/s41388-019-0768-8.

[112] G. Hernández, J.L. Ramírez, A. Pedroza-Torres, L.A. Herrera, M.A. Jiménez-Ríos, The secret life of translation initiation in prostate cancer, Front. Genet. 10 (2019) 1–10. https://doi.org/10.3389/fgene.2019.00014.

[113] H. Ziouziou, C. Andrieu, E. Laurini, S. Karaki, M. Fermeglia, R. Oueslati, D. Taieb, M. Camplo, O. Siri, S. Pricl, M. Katsogiannou, P. Rocchi, Targeting Hsp27/eIF4E interaction with phenazine compound: A promising alternative for castration-resistant prostate cancer treatment, Oncotarget. 8 (2017) 77317–77329. https://doi.org/10.18632/oncotarget.20469.

[114] Y. Frosi, R. Usher, D.T.G. Lian, D.P. Lane, C.J. Brown, Monitoring flux in signalling pathways through measurements of 4EBP1-mediated eIF4F complex assembly, BMC Biol. 17 (2019) 1–19. https://doi.org/10.1186/s12915-019-0658-0.

[115] F.Y. Feng, V. Kothari, Driven to metastasize: Kinases as potential therapeutic targets in prostate cancer, Proc. Natl. Acad. Sci. U. S. A. 113 (2016) 473–475. https://doi.org/10.1073/pnas.1522938113.

[116] J. Stebbing, L.C. Lit, H. Zhang, R.S. Darrington, O. Melaiu, B. Rudraraju, G. Giamas, The regulatory roles of phosphatases in cancer, Oncogene. 33 (2014) 939–953. https://doi.org/10.1038/onc.2013.80.

[117] H. Whitworth, S. Bhadel, M. Ivey, M. Conaway, A. Spencer, R. Hernan, H. Holemon, D. Gioeli, Identification of kinases regulating prostate cancer cell growth using an RNAi phenotypic screen, PLoS One. 7 (2012) 1–11. https://doi.org/10.1371/journal.pone.0038950.

[118] R.R. Ruela-De-Sousa, E. Hoekstra, A. Marije Hoogland, K.C. Souza Queiroz, M.P. Peppelenbosch, A.P. Stubbs, K. Pelizzaro-Rocha, G.J.L.H. Van Leenders, G. Jenster, H. Aoyama, C. V. Ferreira, G.M. Fuhler, Low-molecular-weight protein tyrosine phosphatase predicts prostate cancer outcome by increasing the metastatic potential, Eur. Urol. 69 (2016) 710–719. https://doi.org/10.1016/j.eururo.2015.06.040.

[119] C.E. Nunes-Xavier, J. Mingo, J.I. López, R. Pulido, The role of protein tyrosine phosphatases in prostate cancer biology, Biochim. Biophys. Acta - Mol. Cell Res. 1866 (2019) 102–113. https://doi.org/10.1016/j.bbamcr.2018.06.016.

[120] T.C. Yuan, F.F. Lin, S. Veeramani, S.J. Chen, H.S. Earp, M.F. Lin, ErbB-2 via PYK2 upregulates the adhesive ability of androgen receptor-positive human prostate cancer cells, Oncogene. 26 (2007) 7552–7559. https://doi.org/10.1038/sj.onc.1210570.

[121] S. Agarwal, K.S. Amin, S. Jagadeesh, G. Baishay, P.G. Rao, N.C. Barua, S. Bhattacharya, P.P. Banerjee, Mahanine restores RASSF1A expression by down-regulating DNMT1 and DNMT3B in prostate cancer cells, Mol. Cancer. 12 (2013) 1–12. https://doi.org/10.1186/1476-4598-12-99.

[122] Y.N. Xue, B.B. Yu, J.L. Li, R. Guo, L.C. Zhang, L.K. Sun, Y.N. Liu, Y. Li, Zinc and p53 disrupt mitochondrial binding of HK2 by phosphorylating VDAC1, Exp. Cell Res. 374 (2019) 249–258. https://doi.org/10.1016/j.yexcr.2018.12.002.

[123] U. Demir, A. Koehler, R. Schneider, S. Schweiger, H. Klocker, Metformin anti-tumor effect via disruption of the MID1 translational regulator complex and AR downregulation in prostate cancer cells, BMC Cancer. 14 (2014) 1–9. https://doi.org/10.1186/1471-2407-14-52.

[124] S.Z. Zlivianski, M.S. Pellman, M.K. Ellerman, V.K. Akitelashvilli, X.Z. Hou, E.L. Ugo, M.L. Ee, R.T. Aylor, T.L.D. Avis, R.H. Auke, M.L. In, ERK INHIBITOR PD98059 ENHANCES DOCETAXEL-INDUCED APOPTOSIS OF ANDROGEN-INDEPENDENT HUMAN PROSTATE CANCER CELLS, 485 (2003) 478–485. https://doi.org/10.1002/ijc.11413.

[125] K. Amin, S. Jagadeesh, G. Baishya, P. Rao, A Naturally-Derived Small Molecule Disrupts Ligand-Dependent and Ligand-Independent Androgen Receptor Signaling in Human Prostate Cancer Cells Karishma, 13 (2015) 341–352. https://doi.org/10.1158/1535-7163.MCT-13-0478.A.

[126] D. Dutta, R. Das, C. Mandal, C. Mandal, Structure-Based Kinase Pro fi ling To Understand the Polypharmacological Behavior of Therapeutic Molecules, (2018). https://doi.org/10.1021/acs.jcim.7b00227.

[127] C. Luis, N. Doctor, M. Surgeon, M. Oncologist, Metabolism of the prostate and progression to Prostate Cancer, (n.d.).

[128] P. Pandey, P. Seshacharyulu, S. Das, S. Rachagani, M.P. Ponnusamy, Y. Yan, S.L. Johansson, K. Datta, M.F. Lin, S.K. Batra, Impaired expression of protein phosphatase 2A subunits enhances metastatic potential of human prostate cancer cells through activation of AKT pathway, Br. J. Cancer. 108 (2013) 2590–2600. https://doi.org/10.1038/bjc.2013.160.

[129] L. Beatriz, A. Rojas, M.B. Gomes, Metformin : an old but still the best treatment for type 2 diabetes, (2013) 1–15.

[130] Y. Lei, Y. Yi, Y. Liu, X. Liu, E.T. Keller, C.N. Qian, J. Zhang, Y. Lu, Metformin targets multiple signaling pathways in cancer, (2017) 1–9. https://doi.org/10.1186/s40880-017-0184-9.

[131] C. Rothermundt, S. Hayoz, A.J. Templeton, R. Winterhalder, R.T. Strebel, D. Bärtschi, M. Pollak, L. Lui, K. Endt, R. Schiess, J.H. Rüschoff, R. Cathomas, S. Gillessen, Metformin in chemotherapy-naive castration-resistant prostate cancer: A multicenter phase 2 trial (SAKK 08/09), Eur. Urol. 66 (2014) 468–474. https://doi.org/10.1016/j.eururo.2013.12.057.

[132] B. Gui, F. Gui, T. Takai, C. Feng, X. Bai, L. Fazli, X. Dong, S. Liu, X. Zhang, W. Zhang, A.S.

Kibel, L. Jia, Selective targeting of PARP-2 inhibits androgen receptor signaling and prostate cancer growth through disruption of FOXA1 function, Proc. Natl. Acad. Sci. U. S. A. 116 (2019) 14573–14582. https://doi.org/10.1073/pnas.1908547116.

[133] H. Cheng, X. Li, C. Wang, Y. Chen, S. Li, J. Tan, B. Tan, Y. He, Inhibition of tankyrase by a novel small molecule significantly attenuates prostate cancer cell proliferation, Cancer Lett. 443 (2019) 80–90. https://doi.org/10.1016/j.canlet.2018.11.013.

[134] E. Kounatidou, S. Nakjang, S.R.C. McCracken, S.M. Dehm, C.N. Robson, D. Jones, L. Gaughan, A novel CRISPR-engineered prostate cancer cell line defines the AR-V transcriptome and identifies PARP inhibitor sensitivities, Nucleic Acids Res. 47 (2019) 5634–5647. https://doi.org/10.1093/nar/gkz286.

[135] V. Virtanen, K. Paunu, J.K. Ahlskog, R. Varnai, C. Sipeky, M. Sundvall, PARP inhibitors in prostate cancer—The preclinical rationale and current clinical development, Genes (Basel). 10 (2019) 1–19. https://doi.org/10.3390/genes10080565.

[136] I. Ahmad, O.J. Sansom, Role of Wnt signalling in advanced prostate cancer, J. Pathol. 245 (2018) 3–5. https://doi.org/10.1002/path.5029.

[137] Y. Dai, M. Liu, W. Tang, J. Desano, E. Burstein, M. Davis, K. Pienta, T. Lawrence, L. Xu, Molecularly targeted radiosensitization of human prostate cancer by modulating inhibitor of apoptosis, Clin. Cancer Res. 14 (2008) 7701–7710. https://doi.org/10.1158/1078-0432.CCR-08-0188.

[138] L. Yin, Y. Liu, Y. Peng, Y. Peng, X. Yu, Y. Gao, B. Yuan, Q. Zhu, T. Cao, L. He, Z. Gong, L. Sun, X. Fan, X. Li, PARP inhibitor veliparib and HDAC inhibitor SAHA synergistically co-target the UHRF1/BRCA1 DNA damage repair complex in prostate cancer cells, J. Exp. Clin. Cancer Res. 37 (2018) 1–14. https://doi.org/10.1186/s13046-018-0810-7.

[139] A.B. Pilling, O. Hwang, A. Boudreault, A. Laurent, C. Hwang, IAP Antagonists Enhance Apoptotic Response to Enzalutamide in Castration-Resistant Prostate Cancer Cells via Autocrine TNF- a Signaling, (2017). https://doi.org/10.1002/pros.23327.

[140] C.J. Norbury, B. Zhivotovsky, DNA damage-induced apoptosis, Oncogene. 23 (2004) 2797–2808. https://doi.org/10.1038/sj.onc.1207532.

[141] P.R. Geethakumari, M.J. Schiewer, K.E. Knudsen, W.K. Kelly, PARP Inhibitors in Prostate Cancer, (2017). https://doi.org/10.1007/s11864-017-0480-2.

[142] H. Lu, L. Bai, Y. Zhou, Recent Study of Dual HDAC/PARP Inhibitor for the Treatment of Tumor, Curr. Top. Med. Chem. 19 (2019). https://doi.org/10.2174/1568026619666190603092407.

[143] B. AL-Husein, A. Goc, P.R. Somanath, Suppression of interactions between prostate tumor cell-surface integrin and endothelial ICAM-1 by simvastatin inhibits micrometastasis, J. Cell. Physiol. 228 (2013) 2139–2148. https://doi.org/10.1002/jcp.24381.

[144] A.E. Dart, D.C. Worth, G. Muir, A. Chandra, J.D. Morris, C. McKee, C. Verrill, R.J. Bryant, P.R. Gordon-Weeks, The drebrin/EB3 pathway drives invasive activity in prostate cancer, Oncogene. 36 (2017) 4111–4123. https://doi.org/10.1038/onc.2017.45.

[145] Y. Gao, L.U.O. Li, T. Li, L.E.I. Ma, M. Yuan, W.E.I. Sun, H.L.I.N. Cheng, L. Niu, Z. Du, Z. Quan, Y. Fan, J. Fan, C. Luo, X. Wu, Simvastatin delays castration - resistant prostate cancer metastasis and androgen receptor antagonist resistance by regulating the expression of caveolin - 1, (2019) 2054–2068. https://doi.org/10.3892/ijo.2019.4774.

[146] M. Schonewille, J.F. De Boer, L. Mele, H. Wolters, V.W. Bloks, J.C. Wolters, J.A. Kuivenhoven, U.J.F. Tietge, G. Brufau, A.K. Groen, Statins increase hepatic cholesterol synthesis and stimulate fecal cholesterol elimination in mice, 57 (2016). https://doi.org/10.1194/jlr.M067488.

[147] K. Pelton, M. Freeman, K. Solomon, Cholesterol and Prostate Cancer Kristine, 12 (2013) 751–759. https://doi.org/10.1016/j.coph.2012.07.006.Cholesterol.

[148] S.K. Martin, M. Kamelgarn, N. Kyprianou, Cytoskeleton targeting value in prostate cancer treatment., Am. J. Clin. Exp. Urol. 2 (2014) 15–26. http://www.ncbi.nlm.nih.gov/pubmed/25374905%0Ahttp://www.pubmedcentral.nih.gov/articlerender.fcgi?artid=PMC4219288.

[149] G.M. Nitulescu, C. Draghici, O.T. Olaru, L. Matei, A. Ioana, L.D. Dragu, C. Bleotu, Synthesis and apoptotic activity of new pyrazole derivatives in cancer cell lines, Bioorg. Med. Chem. (2015). https://doi.org/10.1016/j.bmc.2015.07.010.

[150] A.D. Cunningham, N. Qvit, D. Mochly-rosen, Peptides and peptidomimetics as regulators of protein-protein interactions, (2018) 59–66. https://doi.org/10.1016/j.sbi.2016.12.009.Peptides.

[151] H. Bruzzoni-Giovanelli, V. Alezra, N. Wolff, C.Z. Dong, P. Tuffery, A. Rebollo, Interfering peptides targeting protein–protein interactions: the next generation of drugs?, Drug Discov. Today. 23 (2018) 272–285. https://doi.org/10.1016/j.drudis.2017.10.016.

[152] J. Chatterjee, M. Beullens, R. Sukackaite, J. Qian, B. Lesage, D.J. Hart, M. Bollen, M. Kçhn, Angewandte Development of a Peptide that Selectively Activates Protein Phosphatase-1 in Living Cells **, (2012) 10054–10059. https://doi.org/10.1002/anie.201204308.

[153] J.L. Lau, M.K. Dunn, Bioorganic & Medicinal Chemistry Therapeutic peptides : Historical perspectives , current development trends , and future directions, Bioorg. Med. Chem. 26 (2018) 2700–2707. https://doi.org/10.1016/j.bmc.2017.06.052.

[154] W. Gao, Androgen receptor as a therapeutic target ☆, Adv. Drug Deliv. Rev. 62 (2010) 1277–1284. https://doi.org/10.1016/j.addr.2010.08.002.

[155] M. Nakka, I.U. Agoulnik, N.L. Weigel, Targeted disruption of the p160 coactivator interface of androgen receptor (AR) selectively inhibits AR activity in both androgen-dependent and castration-resistant AR-expressing prostate cancer cells, Int. J. Biochem. Cell Biol. 45 (2013) 763–772. https://doi.org/10.1016/j.biocel.2012.12.012.

[156] P. Ravindranathan, T.K. Lee, L. Yang, M.M. Centenera, L. Butler, W.D. Tilley, J.T. Hsieh, J.M. Ahn, G. V. Raj, Peptidomimetic targeting of critical androgen receptor-coregulator interactions in prostate cancer, Nat. Commun. 4 (2013) 1911–1923. https://doi.org/10.1038/ncomms2912.

[157] K. Nishimura, H.-J. Ting, Y. Harada, T. Tokizane, N. Nonomura, H.-Y. Kang, H.-C. Chang, S. Yeh, H. Miyamoto, M. Shin, K. Aozasa, A. Okuyama, C. Chang, Modulation of androgen receptor transactivation by gelsolin: a newly identified androgen receptor coregulator., Cancer Res. 63 (2003) 4888–94. https://doi.org/10.1074/jbc.M506147200.

[158] K.A. Link, S. Balasubramaniam, A. Sharma, C.E.S. Comstock, S. Godoy-Tundidor, N.

Powers, K.H. Cao, A. Haelens, F. Claessens, M.P. Revelo, K.E. Knudsen, Targeting the BAF57 SWI/SNF subunit in prostate cancer: A novel platform to control androgen receptor activity, Cancer Res. 68 (2008) 4551–4558. https://doi.org/10.1158/0008-5472.CAN-07-6392.

[159] A. Migliaccio, L. Varricchio, A. De Falco, G. Castoria, C. Arra, H. Yamaguchi, A. Ciociola, M. Lombardi, R. Di Stasio, A. Barbieri, A. Baldi, M. V. Barone, E. Appella, F. Auricchio, Inhibition of the SH3 domain-mediated binding of Src to the androgen receptor and its effect on tumor growth, Oncogene. 26 (2007) 6619–6629. https://doi.org/10.1038/sj.onc.1210487.

[160] G. Kuser-Abali, A. Alptekin, M. Lewis, I.P. Garraway, B. Cinar, YAP1 and AR interactions contribute to the switch from androgen-dependent to castration-resistant growth in prostate cancer, Nat. Commun. 6 (2015) 1–13. https://doi.org/10.1038/ncomms9126.

[161] M. Goldenberg-Furmanov, I. Stein, E. Pikarsky, H. Rubin, S. Kasem, M. Wygoda, I. Weinstein, H. Reuveni, S.A. Ben-Sasson, Lyn Is a Target Gene for Prostate Cancer: Sequence-Based Inhibition Induces Regression of Human Tumor Xenografts, Cancer Res. 64 (2004) 1058–1066. https://doi.org/10.1158/0008-5472.CAN-03-2420.

[162] M. Carotenuto, P. De Antonellis, C.M. Chiarolla, C. Attanasio, V. Damiani, I. Boffa, N. Aiese, E. Pedone, B. Accordi, G. Basso, L. Navas, C. Imbimbo, M. Zollo, A therapeutic approach to treat prostate cancer by targeting Nm23-H1/h-Prune interaction, Naunyn. Schmiedebergs. Arch. Pharmacol. 388 (2014) 257–269. https://doi.org/10.1007/s00210-014-1035-8.

[163] C. Pan, X. Jin, Y. Zhao, Y. Pan, J. Yang, R.J. Karnes, J. Zhang, L. Wang, H. Huang, AKT - phosphorylated FOXO 1 suppresses ERK activation and chemoresistance by disrupting IQGAP 1- MAPK interaction , EMBO J. 36 (2017) 995–1010. https://doi.org/10.15252/embj.201695534.

[164] F.A. Mercurio, C. Di Natale, L. Pirone, R. Iannitti, D. Marasco, E.M. Pedone, R. Palumbo, M. Leone, The Sam-Sam interaction between Ship2 and the EphA2 receptor: Design and analysis of peptide inhibitors, Sci. Rep. 7 (2017) 1–11. https://doi.org/10.1038/s41598-017-17684-5.

[165] L.S. Lyons, S. Rao, W. Balkan, J. Faysal, C.A. Maiorino, K.L. Burnstein, Ligand-independent activation of androgen receptors by Rho GTPase signaling in prostate cancer, Mol. Endocrinol. 22 (2008) 597–608. https://doi.org/10.1210/me.2007-0158.

[166] F. Wu, S.O. Peacock, S. Rao, S.K. Lemmon, K.L. Burnstein, Novel interaction between the co-chaperone cdc37 and rho gtpase exchange factor vav3 promotes androgen receptor activity and prostate cancer growth, J. Biol. Chem. 288 (2013) 5463–5474. https://doi.org/10.1074/jbc.M112.390963.

[167] J.A. Schneider, T.W. Craven, A.C. Kasper, C. Yun, M. Haugbro, E.M. Briggs, V. Svetlov, E. Nudler, H. Knaut, R. Bonneau, M.J. Garabedian, K. Kirshenbaum, S.K. Logan, Design of Peptoid-peptide Macrocycles to Inhibit the β-catenin TCF Interaction in Prostate Cancer, Nat. Commun. 9 (2018) 1–10. https://doi.org/10.1038/s41467-018-06845-3.

[168] P.K. Sharma, R. Singh, K.R. Novakovic, J.W. Eaton, W.E. Grizzle, S. Singh, CCR9 mediates PI3K/AKT-dependent antiapoptotic signals in prostate cancer cells and inhibition of CCR9-CCL25 interaction enhances the cytotoxic effects of etoposide, Int. J. Cancer. 127 (2010) 2020–2030. https://doi.org/10.1002/ijc.25219.

[169] Y. Teng, A. Bahassan, D. Dong, L.E. Hanold, X. Ren, E.J. Kennedy, J.K. Cowell, Targeting the WASF3-CYFIP1 complex using stapled peptides suppresses cancer cell invasion, Cancer Res. 76 (2016) 965–973. https://doi.org/10.1158/0008-5472.CAN-15-1680.

[170] D.S. Aaronson, M. Muller, S.R. Neves, W.C. Chung, G. Jayaram, R. Iyengar, P.T. Ram, An androgen-IL-6-Stat3 autocrine loop re-routes EGF signal in prostate cancer cells, Mol. Cell. Endocrinol. 270 (2007) 50–56. https://doi.org/10.1016/j.mce.2007.02.006.

[171] S.Y. Park, X. Yu, C. Ip, J.L. Mohler, P.N. Bogner, Y.M. Park, Peroxiredoxin 1 interacts with androgen receptor and enhances its transactivation, Cancer Res. 67 (2007) 9294–9303. https://doi.org/10.1158/0008-5472.CAN-07-0651.

[172] H. Wang, N.C. McKnight, T. Zhang, M.L. Lu, S.P. Balk, X. Yuan, SOX9 is expressed in normal prostate basal cells and regulates androgen receptor expression in prostate cancer cells, Cancer Res. 67 (2007) 528–536. https://doi.org/10.1158/0008-5472.CAN-06-1672.

[173] Z. Wang, Z. Wang, J. Guo, Y. Li, J.H. Bavarva, C. Qian, M.C. Brahimi-Horn, D. Tan, W. Liu,

Inactivation of androgen-induced regulator ARD1 inhibits androgen receptor acetylation and prostate tumorigenesis, Proc. Natl. Acad. Sci. U. S. A. 109 (2012) 3053–3058. https://doi.org/10.1073/pnas.1113356109.

[174] H. Ishiguro, K. Izumi, Y. Li, Y. Zheng, E. Kashiwagi, T. Kawahara, H. Miyamoto, Mp31-08 Semenogelin I Promotes Prostate Cancer Cell Growth Via Functioning As an Androgen Receptor Coactivator and Protecting Against Zinc Cytotoxicity, J. Urol. 191 (2014) 738–747. https://doi.org/10.1016/j.juro.2014.02.917.

[175] S. Chen, C.T. Kesler, B.M. Paschal, S.P. Balk, Androgen receptor phosphorylation and activity are regulated by an association with protein phosphatase 1, J. Biol. Chem. 284 (2009) 25576–25584. https://doi.org/10.1074/jbc.M109.043133.

[176] X. Liu, W. Han, S. Gulla, N.I. Simon, Y. Gao, C. Cai, H. Yang, X. Zhang, J. Liu, S.P. Balk, S. Chen, Protein phosphatase 1 suppresses androgen receptor ubiquitylation and degradation, Oncotarget. 7 (2016) 1754–1764. https://doi.org/10.18632/oncotarget.6434.

[177] P. Leister, A. Felten, A.I. Chasan, K.H. Scheidtmann, ZIP kinase plays a crucial role in androgen receptor-mediated transcription, Oncogene. 27 (2008) 3292–3300. https://doi.org/10.1038/sj.onc.1210995.

[178] L. Li, C.H. Ren, S.A. Tahir, C. Ren, T.C. Thompson, Caveolin-1 Maintains Activated Akt in Prostate Cancer Cells through Scaffolding Domain Binding Site Interactions with and Inhibition of Serine/Threonine Protein Phosphatases PP1 and PP2A, Mol. Cell. Biol. 23 (2003) 9389–9404. https://doi.org/10.1128/mcb.23.24.9389-9404.2003.

[179] O. Pasder, S. Shpungin, Y. Salem, A. Makovsky, S. Vilchick, S. Michaeli, H. Malovani, U. Nir, Downregulation of Fer induces PP1 activation and cell-cycle arrest in malignant cells, Oncogene. 25 (2006) 4194–4206. https://doi.org/10.1038/sj.onc.1209695.

[180] C. Martin-Granados, A.R. Prescott, N. van Dessel, A. van Eynde, M. Arocena, I.P. Klaska, J. Görnemann, M. Beullens, M. Bollen, J. V. Forrester, C.D. McCaig, A role for PP1/NIPP1 in steering migration of human cancer cells, PLoS One. 7 (2012) 1–14. https://doi.org/10.1371/journal.pone.0040769.

[181] A. Ren, G. Yan, B. You, J. Sun, Down-regulation of mammalian sterile 20-like kinase 1

by heat shock protein 70 mediates cisplatin resistance in prostate cancer cells, Cancer Res. 68 (2008) 2266–2274. https://doi.org/10.1158/0008-5472.CAN-07-6248.

[182] J. Villar, H.S. Quadri, I. Song, Y. Tomita, O.M. Tirado, V. Notario, PCPH/ENTPD5 expression confers to prostate cancer cells resistance against cisplatin-induced apoptosis through protein kinase Cα-mediated Bcl-2 stabilization, Cancer Res. 69 (2009) 102–110. https://doi.org/10.1158/0008-5472.CAN-08-2922.

[183] L. BARKI-HARRINGTON, A.L. BOOKOUT, G. WANG, M.E. LAMB, L.M.F. LEEB-LUNDBERG, Y. DAAKA, Requirement for direct cross-talk between B1 and B2 kinin receptors for the proliferation of androgen-insensitive prostate cancer PC3 cells, Biochem. J. 371 (2003) 581–587. https://doi.org/10.1042/bj20021708.

[184] E. Källberg, T. Vogl, D. Liberg, A. Olsson, P. Björk, P. Wikström, A. Bergh, J. Roth, F. Ivars, T. Leanderson, S100A9 Interaction with TLR4 Promotes Tumor Growth, PLoS One. 7 (2012) e34207. https://doi.org/10.1371/journal.pone.0034207.

[185] K.I. Takayama, T. Suzuki, T. Tanaka, T. Fujimura, S. Takahashi, T. Urano, K. Ikeda, S. Inoue, TRIM25 enhances cell growth and cell survival by modulating p53 signals via interaction with G3BP2 in prostate cancer, Oncogene. 37 (2018) 2165–2180. https://doi.org/10.1038/s41388-017-0095-x.

[186] X. Zhang, J. Akech, G. Browne, S. Russell, J.J. Wixted, J.L. Stein, G.S. Stein, J.B. Lian, Runx2-smad signaling impacts the progression of tumor-induced bone disease, Int. J. Cancer. 136 (2015) 1321–1332. https://doi.org/10.1002/ijc.29094.

[187] S. Das, P. Shetty, M. Valapala, S. Dasgupta, Z. Gryczynski, J.K. Vishwanatha, Signal transducer and activator of transcription 6 (STAT6) Is a novel interactor of annexin A2 in prostate cancer cells, Biochemistry. 49 (2010) 2216–2226. https://doi.org/10.1021/bi9013038.

[188] C. Chen, Q. Zhang, S. Liu, K.R. Parajuli, Y. Qu, J. Mei, Z. Chen, H. Zhang, D.B. Khismatullin, Z. You, IL-17 and insulin/IGF1 enhance adhesion of prostate cancer cells to vascular endothelial cells through CD44-VCAM-1 interaction, Prostate. 75 (2015) 883–895. https://doi.org/10.1002/pros.22971.

[189] L. Yu, Z.F. Shang, J. Wang, H. Wang, F. Huang, Z. Zhang, Y. Wang, J. Zhou, S. Li, PC-1/PrLZ

confers resistance to rapamycin in prostate cancer cells through increased 4E-BP1 stability, Oncotarget. 6 (2015) 20356–20369. https://doi.org/10.18632/oncotarget.3931.

**Table 1**: Summary of the small molecules used to the disruption of protein-protein interactions critical for progression of prostate cancer. The respective output of the protein interactions blockage *in vitro* and *in vivo* and clinical trials (when available) are also included.

| | | | ***In vitro*** | | **Pre-clinical validation** | | | **Clinical trial** | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **Small molecule** | **Type of PPI** | **Targeted PPI** | **Cell line** | **Output of disruption *in vitro*** | **Tested in vivo** | **Output of disruption *in vivo*** | **Refs** | **Clinical trial** | **Output** | **Refs.** | **Selectivity** | **Application in CRPC treatment** |
| PTER-ITC conjugate | Androgen receptor (AR)-related | AR/ SRC1, AR/ GRIP1 | LNCaP, PC-3 | ↓ PCa cells proliferation and induce apoptosis | - | - | [52] | - | - | - | No data | - |
| iCRT3 | | βcatenin/ AR | LNCaP | ↓ PCa cells growth | mice (xenografts) | inhibited tumor growth | [53] | - | - | - | No data | Potential role |
| Ailanthone | | AR/HSP90 | LNCaP, 22RV1 | blocks tumor growth and metastasis | mice (xenografts) | inhibited tumor growth | [56] | - | - | - | No data | Described role |
| OGX-427 | | AR/ HSP27 | LNCaP | ↑ PCa cells apoptosis | - | - | [57] | Phase I Phase II | Safe and well tolerated; ↓PSA | [58,59] | No data | Described role |
| Roscovitine | | Cdk5/ AR | LNCaP | inhibit PCa cells proliferation | - | - | [61] | - | - | - | Pan-CDK inhibitor | - |
| Quercetin | Bcl-2 protein family-related | BclxL/ Bax | LNCaP | ↑ PCa cells apoptosis | - | - | [69] | - | - | - | PAINS | Potential role |
| (--Gossypol | | BclxL/ Bax or Bad | PC-3 | induce apoptosis (mitochondria pathway); enhance antitumor activity of docetaxel | mice (xenografts) | enhance docetaxel- and radiation therapy-induce apoptosis | [71] [72] | Phase I/II | PSA levels↓ in some patients; no objective response (RECIST) | [29] | PAINS | Potential role |
| α-tocopheryl succinate | | BclxL or Bcl2/ Bak | PC-3 | induces PCa cells apoptosis | - | - | [76] | - | - | - | Target multiplicity | - |
| ABT-737 | | Bcl2 /Bax | PC-3, LNCaP | ↑ PCa cells apoptosis, sensitized PCa cells to docetaxel | - | - | [79] | - | - | - | Higher affinity than other BCl-2 inhibitors | Described role |
| DATS | | Bcl2/ Bax | PC-3 | induces PCa cells apoptosis | mice (xenografts) | inhibited tumor progression and lung metastasis | [82] [83] | - | - | - | Target multiple signaling pathways | - |
| (-)-Gossypol | | Bcl2/ Becn1 | LNCaP | induces PCa cells autophagy | mice (xenografts) | inhibit tumor growth | [86] | - | - | - | PAINS | Potential role |
| 3-AWA | | Bcl-2/Becn1 | PC-3, DU145 | ↑ PCa cells apoptosis | - | - | [87] | - | - | - | Highly selective MMP2 inhibitor | - |

| | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Apigenin | | Bax/Ku70 | PC-3, DU145 | ↑ PCa cells apoptosis | mice (xenografts) | inhibit tumor growth | [91] | - | - | - | PAINS | Potential role |
| | | Bad/ 14-3-3β | PC-3 | induce PCa cells apoptosis | - | - | [92] | - | - | - | | |
| DATS | | Bad/14-3-3β | PC-3, DU145 | induces PCa cells apoptosis | - | - | [93] | - | - | - | Target multiple signaling pathways | - |
| XCT790 | Other transcription/ translation factors-related | ERRα/ HIF1α | LNCaP, PC-3 | ↓ PCa cells proliferation | - | - | [103] | - | - | - | Selective ERRα inverse agonist | - |
| ETPs | | HIF1α/ P300 | PC-3 | ↓ tumor growth and angiogenesis | mice (xenografts) | inhibited tumor growth | [104] | - | - | - | Multiple targets suggested | - |
| NC043 | | βcatenin/ TCF4 | PC-3, LNCaP, DU145 | ↓ PCa cells malignancy, migration | mice (xenografts) | impaired tumorigenesis | [108] | - | - | - | No data | Potential role |
| 10058-F4 | | cMyc/ Max | DU145 PC-3 | Inhibited cMyc/Max | mice (xenografts) | no signif. Inhibition of tumor growth | [111] | - | - | - | Complete selectivity for the interaction | Potential role |
| phenazine | | Hsp27/ eIF4E | PC-3 | ↓ viability and ↑ death of PCa cells in chemo- and castration-resistant PCa | mice (xenografts) | ↓ tumor growth | [115] | - | - | - | Selective | Described role |
| BEZ235 | | eIF4G/ eIF4E | PC-3 | decrease chemoresistance | - | - | [116] | - | - | - | No data | Potential role |
| PD98059 | Kinases/ phosphatases-related | ErbB2/ PYK2 | LNCaP, C-81 | abolish adhesive ability of PCa cells | - | - | [122] | - | - | - | Selective MEK inhibitor | Potential role |
| mahanine | | pAkt/ DNMT1 and DNMT3B | PC-3, LNCaP | ↓ PCa cells proliferation, survival | - | - | [123] | - | - | - | Polypharmacological action | Potential role |
| p53 and zinc | | HK2/ VDAC | DU145, PC-3 | ↑ PCa cells apoptosis | mice (xenografts) | inhibited tumor growth | [124] | - | - | - | No data | - |

| | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Metformin | | MID1-α4/PP2A | LNCaP | ↓ PCa cells proliferation and migration; ↓ AR protein levels | - | - | [125] | Phase II | induce CRPC stabilization | [133] | Multiple targets | Described role |
| UPF 1069 | Other PPIs | PARP2/ FOXA1 | LNCaP, 22RV1, PC3 | ↓ PCa cells growth | - | - | [134] | - | - | - | Selective inhibitor of PARP2 | Described role |
| C44 | | TNKS/ USP25 | PC-3, Lapa | ↓ PCa cells proliferation | mice (xenografts) | inhibit PCa cells proliferation | [135] | - | - | - | Selective | Potential role |
| SH-130 | | XIAP/ Smac | DU145 | enhance radiation-induced apoptosis | mice (xenografts) | sensitize tumors to X-ray radiation | [139] | - | - | - | No data | Described role |
| Veliparib, SAHA | | UHRF1/ BRCA1 | LNCaP, PC-3, DU-145, | ↓ PCa cells colony formation, ↑ PCa apoptosis | mice (xenografts) | inhibit tumor growth | [140] | - | - | - | No data | - |
| Simvastatin | | integrinαv β3/ ICAM1 | PC-3 | inhibit PCa micrometastasis | - | - | [145] | Phase I/II | *undergoing* | NCT00572468 | Target enzyme of cholesterol synthesis | Described role |
| BTP2 | | drebin/ EB3 | PC-3 | ↓ PCa cells invasion | - | - | [146] | - | - | - | No data | - |

**Table 2**: Summary of the peptides used to the disruption of protein-protein interactions critical for progression of prostate cancer. The respective output of the protein interactions blockage *in vitro* and *in vivo* and clinical trials (when available) are also included.

| | | | ***In vitro*** | | **Pre-clinical validation** | | | |
|---|---|---|---|---|---|---|---|---|
| **Peptide** | **Type of PPI** | **Targeted PPI** | **Cell line** | **Output of disruption *in vitro*** | **Tested in vivo** | **Output of disruption *in vivo*** | **Refs.** | **Application in CRPC treatment** |
| SRC1-derived peptide | Androgen receptor (AR)-related | AR/ SRC1 or SRC2 | PC-3, VCaP, C4-2 | inhibit PCa cell proliferation | - | - | [157] | Described role |
| peptidomimetics D2 | | AR/ PELP1 | LNCaP | ↓ PCa cells proliferation | mice (xenografts) | Inhibits tumor growth | [158] | - |
| AR peptides | | AR/GSN | LNCaP, PC-3, DU145 | suppression of GSN-enhanced AR activity | - | - | [159] | - |
| BiPep | | AR/ BAF57 | LNCaP, PC-3, DU145 | ↓ PCa cells proliferation | - | - | [160] | - |
| Peptide (10 aminoacid) | | AR/ SRC | LNCaP | ↓ PCa cells proliferation | mice (xenografts) | inhibits tumor growth | [161] | - |
| MST1 kinase | | YAP1/ AR | C4-2, LNCaP | ↓ PCa cells growth | - | - | [162] | Described role |
| KRX-123 (nonapeptide) | Kinases/ phosphatases-related | Lyn interactions | DU145, PC-3 | inhibit PCa cells proliferation | mice (xenografts) | reduce tumor volume | [163] | Described role |
| CPP | | Nm23H1/ h.prone | PC-3 | ↑ apoptosis of PCa cells | mice (xenografts) | ↓ metastasis formation | [164] | - |
| small FOXO1-derived peptide | | IQGAP1/ MAPK | LNCaP, DU145 | suppression of chemoresistance | - | - | [165] | - |
| Peptide (FITC-TAT-(KRI)3) | | Ship2/ EphA2 | PC-3 | PCa cells necrosis | - | - | [166] | - |
| Fragment of Cdc37 | Other PPIs | Cdc37/ Vav3 | PC-3, LNCaP | ↓ PCa cells proliferation | - | - | [168] | Potential role |
| Peptoids | | TCF/ B-catenin | LNCaP | inhibit PCa cell growth | zebrafish | inhibit Wnt signaling | [169] | Potential role |
| CCL25 neut. antibody | | CCR9/ CCL25 | LNCaP, PC-3 | induce apoptosis | mice (xenografts) | ↑ drug efficacy in refractory PCa | [170] | Described role |
| peptides (WAHM1 and 2) | | WASF3/ CYFIP1 | PC-3, DU145 | ↓ PCa cells invasion | - | - | [171] | - |

**Table 3**: Summary of the protein-protein interactions implicated in the progression of PCa and whose disruption has not yet been tested. The role of the protein interactions in PCa is also described.

| Type of PPI | | PPI | Identified in | Role in PCa | Reference |
|---|---|---|---|---|---|
| | AR-related | AR/ STAT3 | LNCaP (*in vitro*) | enhance AR transcriptional activity | [172] |
| | | AR/ PRDX1 | LNCaP and DU145 (*in vitro*) | ↑ AR transactivation | [173] |
| | | AR/ SOX9 | LNCaP and PC-3 (*in vitro*) | ↑ AR protein expression; promotes PCa cells growth | [174] |
| | | AR/ ARD1 | LNCaP (*in vitro*) and human PCa tissue (*in vivo*) | ↑ AR transactivation; contributes to PCa cells proliferation | [175] |
| | | AR/ SEMG1 | LNCaP (*in vitro*) | ↑ AR transactivation; promotes androgen-mediated PCa progression | [176] |
| Kinases-phosphatases-related | | AR/ PP1 | LNCaP (*in vitro*) | ↓ proteasome-mediated AR degradation; ↑ AR-mediated gene transcription | [177] |
| | | | LNCaP and PC-3 (*in vitro*) | supress AR ubiquitylation and degradation; enhance AR activity | [178] |
| | | AR/ ZIPK | LNCaP and PC-3 (*in vitro*) | ↑ AR-mediated transcription | [179] |
| | | PP1/ CAV1 | LNCaP (*in vitro*) | promotes PCa cells survival | [180] |
| | | PP1/ FER | PC-3 (*in vitro*) | promotes PCa cells cycle progression | [181] |
| | | PP1/ NIPP1 | PC-3 (*in vitro*) | ↑ and direct PCa cells migration | [182] |
| | | MST1/ HSP70 | LNCaP (*in vitro*) | induce PCa cells cisplatin resistance | [183] |
| | | PKC/ PCPH | LNCaP and PC-3 (*in vitro*) | ↑ invasiveness of PCa and resistance to cisplatin-induced apoptosis of PCa cells | [184] |
| Other PPIs | | B1R/ B2R | PC-3 (*in vitro*) | induce PCa cells proliferation | [185] |
| | | S100A9/ TLR4 | TRAMP mice model (*in vivo*) | promotes PCa tumor growth | [186] |
| | | TRIM25/ G3BP2 | LNCaP and 22Rv1 (*in vitro*) | enhances PCa cell survival and growth | [187] |
| | | RUNX2/ SMAD | PC-3 (*in vitro*) | mediates tumor growth and metastasis | [188] |
| | | ANXA2/ STAT6 | LNCaP, DU145 and PC-3 (*in vitro*) | potential implications in PCa progression and metastatic process | [189] |
| | | CD44/ VCAM1 | PC-3, DU-145 (*in vitro*) | mediates the adhesion of PCa cells with vascular endothelial cells | [190] |
| | | PC1/ 4EBP1 | LNCaP and C4-2 (*in vitro*) | enhances PCa cells survival and progression and ↑ chemoresistance | [191] |

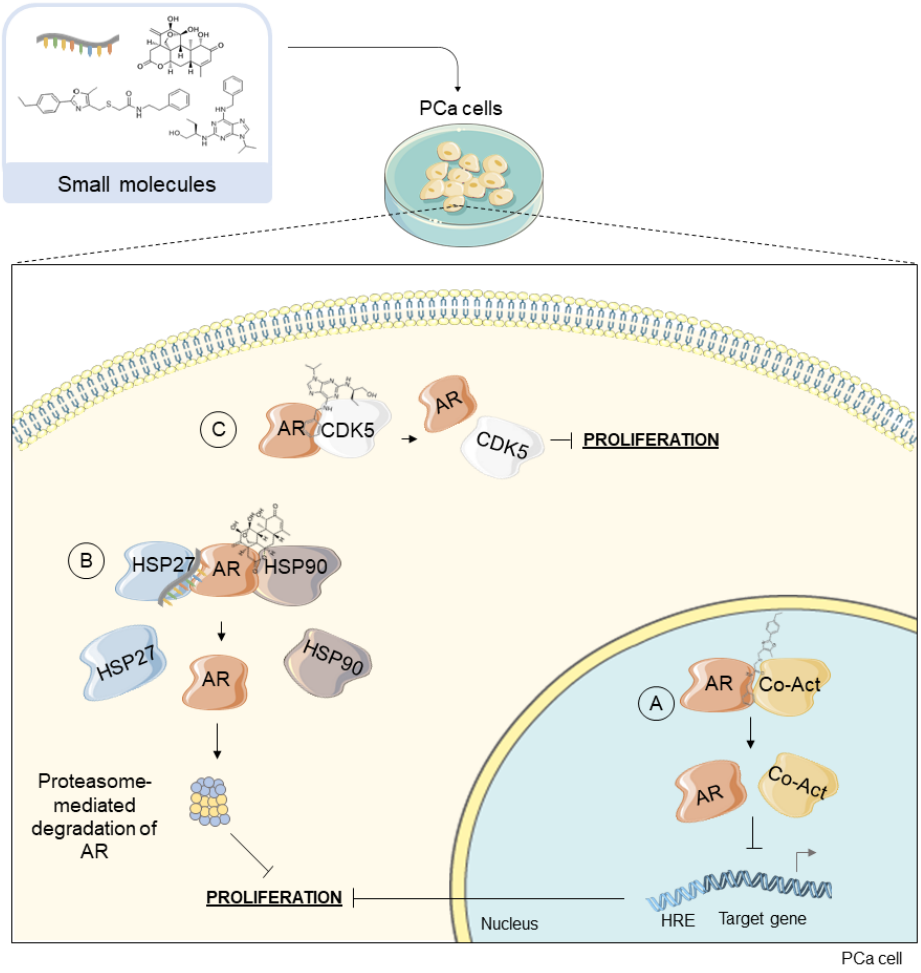


**Figure 1**: **Summary of androgen receptor-related protein-protein interactions, their disruption by small molecules and associated therapeutic outcomes in prostate cancer cells.** A: The disruption of the interaction between AR and its co-activators β-catenin by iCRT3 inhibits AR-mediated gene expression, resulting in decreased PCa cells proliferation. The same effect was observed for the disruption of AR/ SRC1 and GRIP1 by PTER-ITC conjugate; B: The blockage of AR interaction with HSP27 and 90 is achieved by OGX-427 and ailanthone, respectively, resulting in proteasome-degradation of AR, inhibiting its translocation to the nucleus and respective promotion of PCa cells proliferation; C: The disruption of AR/CDK5 interaction by roscovitine leads to reduction of PCa cells proliferation.

Abbreviations: PCa: prostate cancer; AR: Androgen receptor; Co-Act: co-activators; HRE: hormone-response element

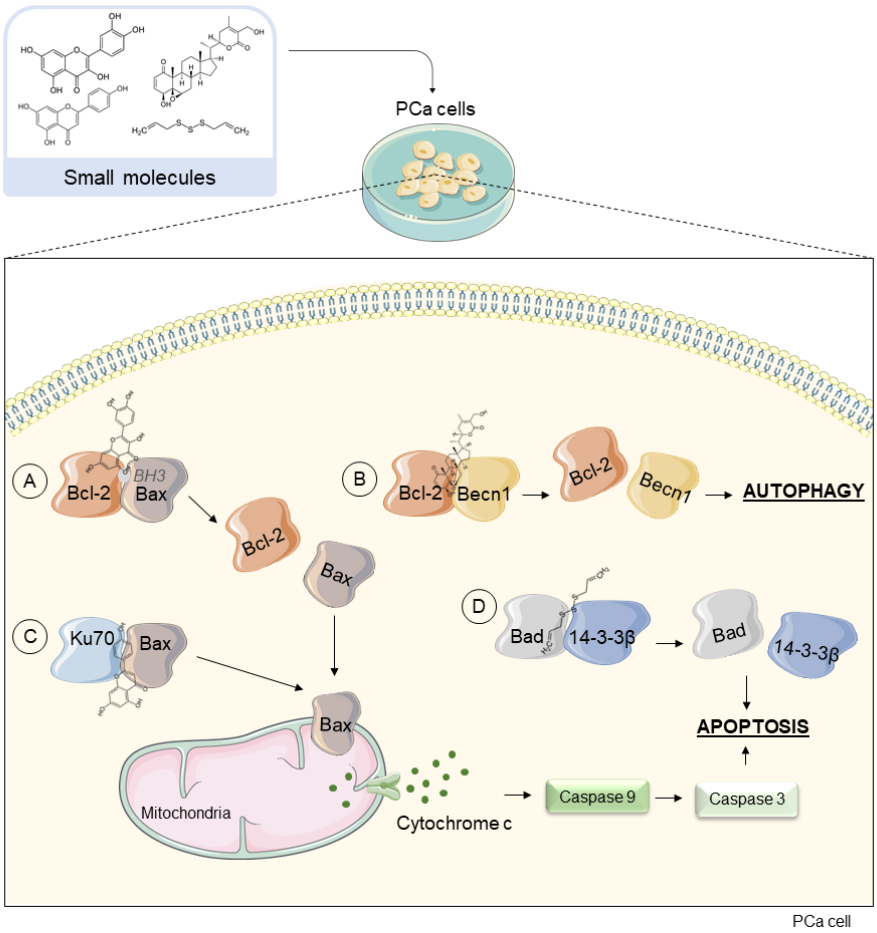


**Figure 2**: **Summary of the Bcl2 protein family-related protein-protein interactions disruption by small molecules and the respective outcomes in prostate cancer cells**. A: The disruption of the interaction between Bcl-2 (or Bcl-xL) with its pro-apoptotic counterparts (Bax, Bad or Bak) by quercetin leads to cytochrome c release from the mitochondria and consequent caspases activation, resulting in PCa cells apoptosis. Other compounds, namely (-)-gossypol, α-tocopheryl-succinate, ABT-737 and diallyl-trisulfide have the same outcome in PCa cells; B: The disruption of the Bcl-2/ Becn1 interaction by 3-AWA results in PCa cells death by autophagy. (-)-gossypol also produces the same effect; C: The interruption of Ku70/ Bax interaction by apigenin releases cytochrome c from the mitochondria, conducing to PCa cells apoptosis; D: The disruption of Bad/ 14-3-3β by diallyl-trisulfide also contributes to PCa cells apoptosis. The same effect is observed with PCa cells incubation with apigenin.

Abbreviations: PCa: prostate cancer

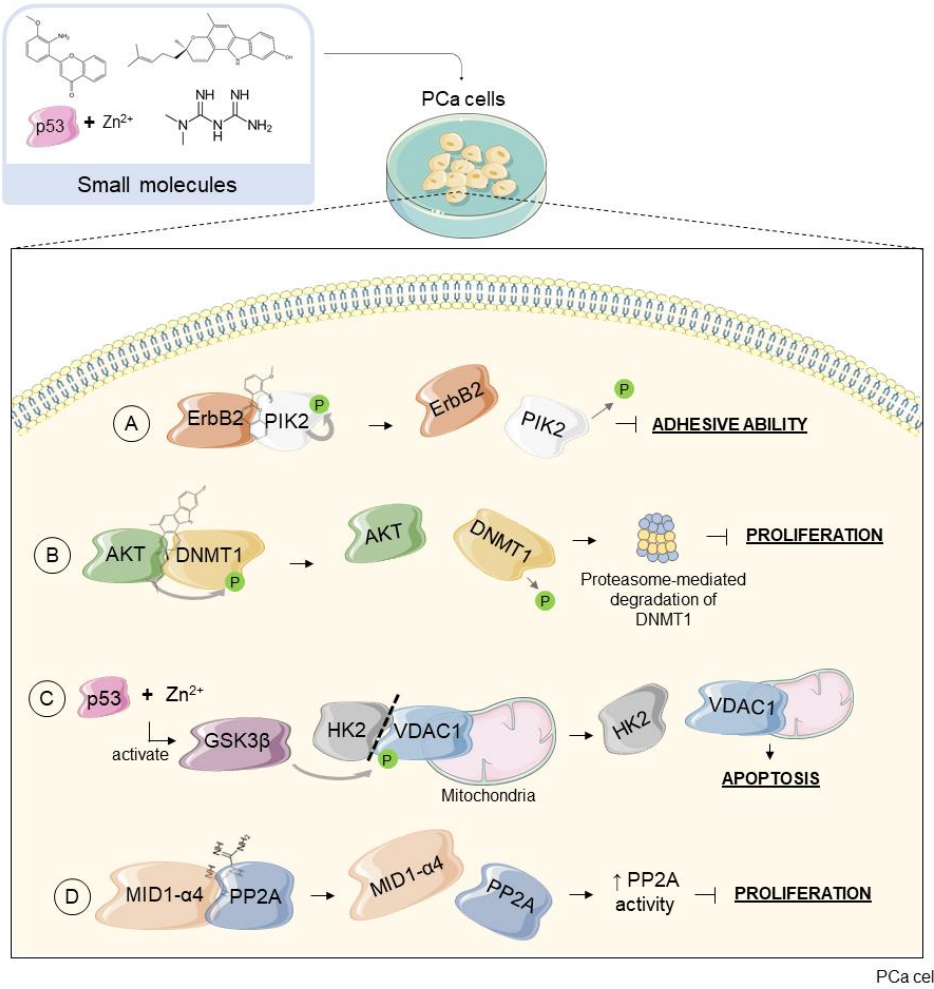


**Figure 3: Summary of the kinases/phosphatases-related protein-protein interactions disruption by small molecules and the respective outcome in prostate cancer cells.** A: The disruption of the interaction between ErbB2 and PIK2 by PD98059 dephosphorylated PIK2 and consequent inhibit PCa cells adhesive ability; B: The blockage of the interaction between DNMT1 and DNMT3B dephosphorylated DNMTs, leading to its proteasome-mediated degradation and consequent inhibition of PCa cells proliferation; C: The zinc, in combination with p53 activates GSK3β, which phosphorylate VDAC1 and consequent inhibits its interaction with HK2 resulting in PCa cells apoptosis; D: The disruption of MID1-α4/PP2A interaction increases the activity of PP2A, inhibiting the proliferation of PCa cells.

Abbreviations: PCa: prostate cancer; P phosphate group

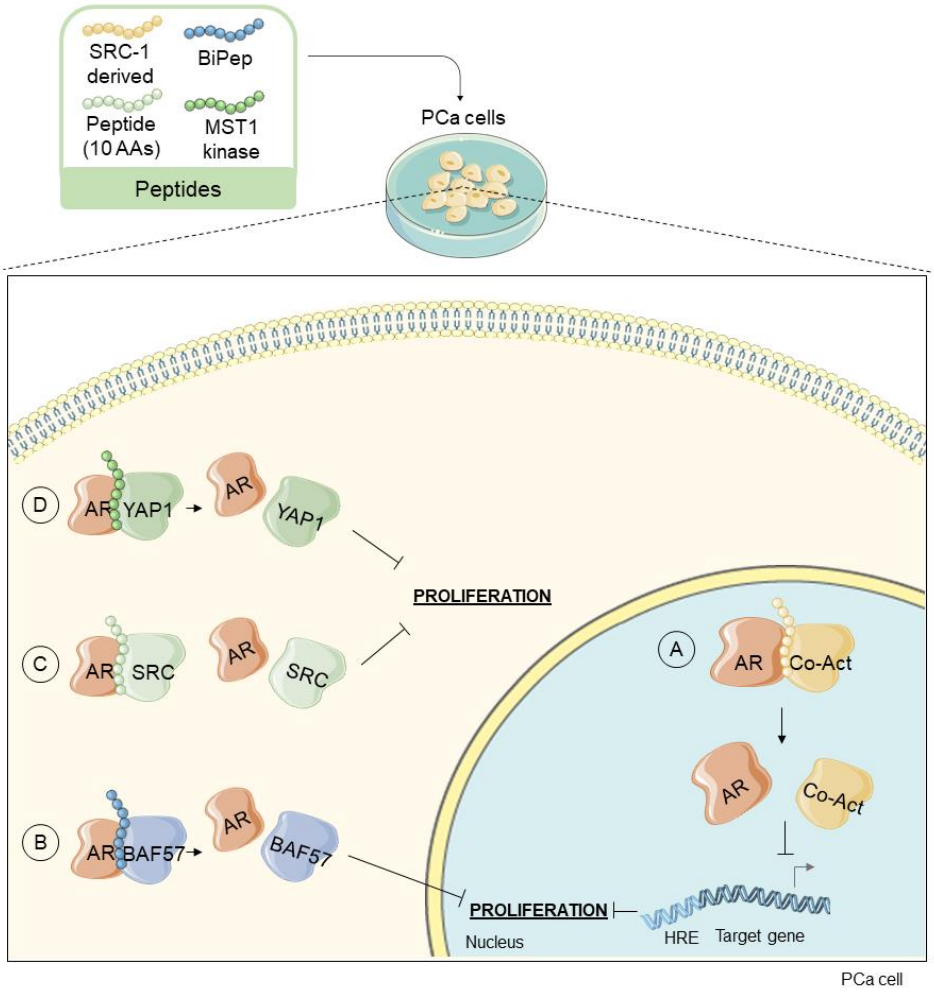


**Figure 4: Summary of androgen receptor-related protein-protein interactions, their disruption by peptides and associated therapeutic outcomes in prostate cancer cells.** A: The disruption of the interaction between AR and its co-activators SRC1 and 2, PELP1 and GSN by peptides based on the protein-protein interactions binding regions - SRC-1 derived peptide, D2 and AR peptides, respectively, inhibits AR transactivation and consequently decrease PCa cells proliferation. B,C: The blockage of the interaction of AR with BAF57 (B) and SRC (C) by an inhibitory peptide of BAF57 and a peptide covering AR-binding motif of SRC, respectively, conduce to reduction of proliferation; D: The interruption of AR/YAP1 interaction by MSTK1 also inhibits PCa cells proliferation.

Abbreviations: PCa: prostate cancer

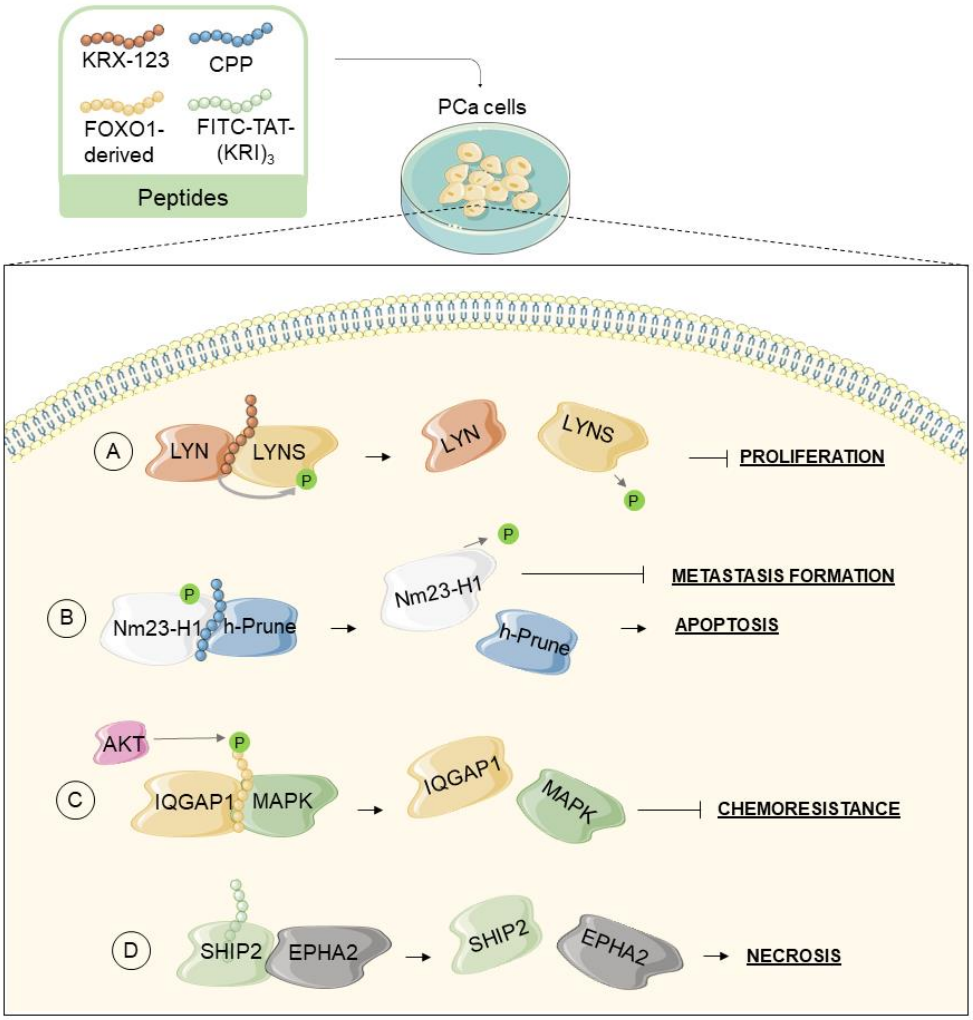


**Figure 5: Summary of kinases/phosphatases-related protein-protein interactions, their disruption by peptides and associated therapeutic outcomes in prostate cancer cells.** A: The disruption of the interaction between LYN and its substrates (LYNS), which are phosphorylated by LYN, by KRX-123 peptide resulted in inhibition of PCa cells proliferation; B: The interruption of Nm23-H1/ h-Prune interaction by a competitive permeable peptide (CPP) dephosphorylated Nm23-H1, leading to inhibition of metastasis and PCa cells apoptosis; C: The blockage of IQGAP1/MAPK interaction by the AKT-phosphorylated FOXO-1 derived peptide conduce to Inhibition of chemoresistance; D: The disruption of SHIP2/ EPHA2 interaction by FITC-TAT-$(KRI)_3$ resulted in PCa cells necrosis.

Abbreviations: PCa: prostate cancer; P phosphate group