

# Chapter 2

## Androgen Action During Prostate Carcinogenesis

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### Abstract

Androgens are critical for normal prostate development and function, as well as prostate cancer initiation and progression. Androgens function mainly by regulating target gene expression through the androgen receptor (AR). Many studies have shown that androgen-AR signaling exerts actions on key events during prostate carcinogenesis. In this review, androgen action in distinct aspects of prostate carcinogenesis, including (i) cell proliferation, (ii) cell apoptosis, and (iii) prostate cancer metastasis will be discussed.

**Key words:** Androgen receptor, prostate cancer, androgen metabolism, androgen signaling, castration-resistant prostate cancer.

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### 1. Androgen Signaling

Androgens are the male sex hormones, which control the differentiation and maturation of male reproductive organs, including the prostate gland. Testosterone is the principal androgen in circulation and is synthesized by Leydig cells in the testes, under the regulation of luteinizing hormone (LH), which is further regulated by gonadotropin-releasing hormone (GnRH). Adrenal glands also synthesize a small amount of androgens, such as dehydroepiandrosterone (DHEA) and androstenedione (4-dione) (1). Testosterone enters prostate cells by passive diffusion, where it is converted enzymatically by 5- $\alpha$  reductases to the more potent androgen dihydrotestosterone (DHT) (2). Binding of androgens to the androgen receptor (AR), a ligand-modulated transcription factor, induces a conformational change in the AR, causing release of heat shock proteins and translocation of the AR to the

nucleus, where it transcriptionally regulates the expression of target genes (3).

In addition to the classic genomic effects of sex steroids, accumulating data have also shown the importance of nongenomic effects (4–7). For instance, androgen treatment results in the association of AR with Src kinase, and thereby activates Src/Raf-1/Erk pathway, leading to cell proliferation and survival (8). Androgens may also post-transcriptionally regulate gene expression by modulating the stability of mRNAs (6). Membrane androgen receptors may also account for some nongenomic effects of androgens (9).

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## 2. Androgen and Prostate Carcinogenesis

Prostate cancer is the most frequently diagnosed cancer and the second leading cause of cancer-related death in US men. The American Cancer Society has estimated that in the USA the number of new cases diagnosed in 2009 was 192,280, and about 27,360 men died of this disease (10). The problem is even more substantial when viewed from a global perspective, with prostate cancer accounting for more than 220,000 deaths worldwide every year (11). Multiple signaling pathways have been demonstrated to be critical for prostate cancer initiation and progression (12, 13), with the androgen signaling pathway being one of the most prominent.

Since the landmark research of Huggins and Hodges in the 1940s, it has been postulated that androgens promote prostate carcinogenesis (14, 15). Although it is well accepted that androgens are critical for prostate cancer growth, it is still controversial whether androgens promote human prostate carcinogenesis *in vivo*. Indeed, there is no increased incidence of prostate cancer in men administered testosterone, there is no reduced risk of prostate cancer in men with low serum androgen levels, and there is no correlation between prostate cancer and serum androgen levels (16, 17). Taken together, these data suggest a ‘saturation’ model of androgen action on androgen-dependent growth (18). This model states that physiologic levels of androgen are important for both normal and malignant prostate cell proliferation, but excessive androgens alone do not lead to uncontrolled cell proliferation. On the other hand, ligand-independent activation of AR signaling plays a critical role in initiation and progression of prostate cancer, particularly following androgen ablation therapy (19, 20). The role of ligand-independent AR activation in prostate carcinogenesis and progression has been discussed in several excellent reviews (1, 21, 22). The

present review focuses on the effects of androgen signaling during critical phases of prostate carcinogenesis.

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### 3. Androgen Action on Prostate Cell Proliferation

Although epidemiologic data suggest that androgens alone are not sufficient to promote prostate carcinogenesis (23), abundant biological data have demonstrated that androgens promote prostate cancer cell proliferation. Androgens induce prostate epithelial cell proliferation via multiple ways, either directly or indirectly.

One of the most common genetic alterations in prostate cancer is the fusion between two genes, i.e., the TMPRSS2 gene and the ETS transcription factor genes, ERG or ETV1 (24). ETS transcription factors are involved in multiple processes, including cell proliferation and cancer cell invasion (25). Current data suggest that up to 72% of all prostate cancers harbor a TMPRSS2-ETS translocation (26–28). TMPRSS2 is a membrane-bound serine protease, which is regulated by androgens and overexpressed in prostate cancers (29, 30). In addition, TMPRSS2 expression is largely limited to prostate, more specifically, prostate luminal epithelial cells (31, 32). The ERG gene is the most commonly overexpressed proto-oncogene in prostate cancer but the underlying mechanism of ERG overexpression was not clear (33). The finding of TMPRSS2-ETS translocations suggests that androgens may promote the expression of ETV1 and ERG, contributing to prostate carcinogenesis. A recent study shows that androgens and irradiation synergistically induce the translocations of TMPRSS2-ERG and TMPRSS2-ETV1 (34). Liganded AR can induce juxtaposition of translocation loci by triggering intra- and inter-chromosomal interactions. Such interactions appear to promote stress-induced site-specific DNA double-stranded breaks at these translocation loci by recruiting activation-induced cytidine deaminase and LINE-1 repeat-encoded ORF2 endonuclease, which are critical for chromosomal translocations. These results suggest a potential mechanism by which androgens promote prostate carcinogenesis through inducing gene translocation.

Increased levels of growth factors, such as insulin-like growth factors (IGFs), fibroblast growth factors (FGFs), and epidermal growth factors (EGFs), are associated with prostate cancer (35–37). Increased expression of growth factors and their receptors promotes prostate cell proliferation, migration, and tumor angiogenesis, thereby facilitating prostate carcinogenesis and cancer progression. IGF-1 has been shown to promote prostate cancer cell proliferation *in vitro* and facilitate the progression of

a prostate cancer xenograft to a castration-recurrent state (35). Androgens regulate the expression of both IGF-1 (38) and its receptor IGF-1R (39) (Fig. 2.1). Two androgen response elements (AREs) exist within the IGF-1 promoter, suggesting that androgens regulate IGF-1 via a direct transcriptional mechanism (38). On the other hand, the effects of androgen on the expression of IGF-1R appear to be through a nongenomic event. Quantitative RT-PCR demonstrated that IGF-1R induction is independent of AR DNA-binding activity. Rather, it appears to depend on the Src/MAPK pathway. Androgen treatment activates Erk1/2 within 5 min, and inhibition of Src/MAPK signaling pathway by various methods can block androgen-induced Erk1/2 activation and IGF-1R expression (39). Another way by which androgens modulate the IGF-1 signaling pathway is through regulation of IGF-binding protein (IGFBP) expression. For example, IGFBP-5 is transcriptionally regulated by androgens (40). Data on androgen regulation on IGFBP-3 are controversial. Some reports have shown that androgens suppress IGFBP-3 levels (41, 42), whereas other studies have reported that androgens increase IGFBP-3 expression (43).

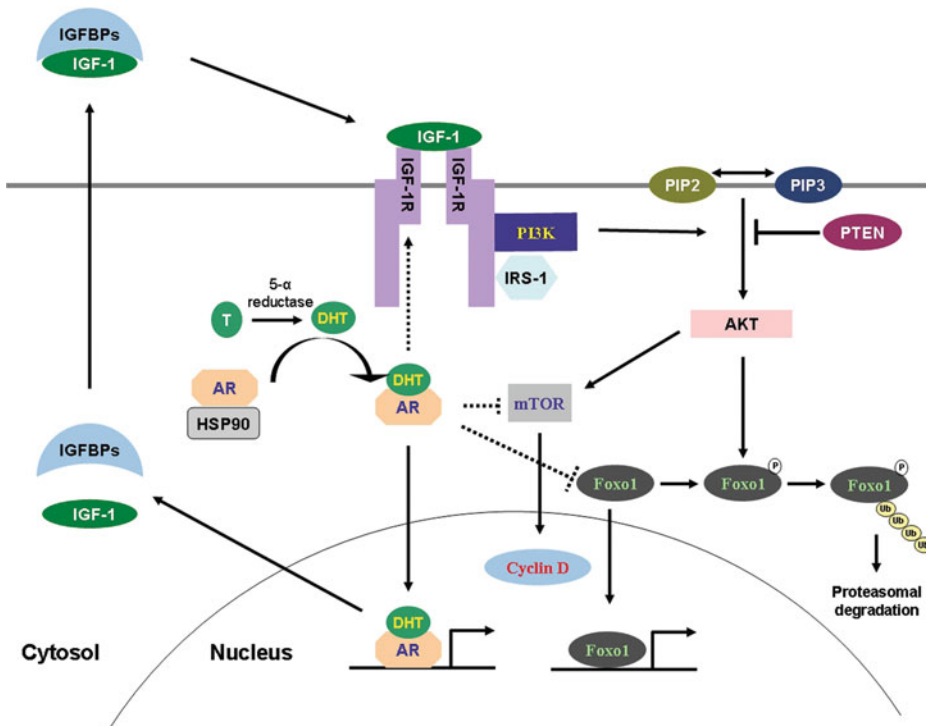


Fig. 2.1. Androgen action on IGF-1 signaling. IGF-1 interacts with IGF-1R to induce PI3K/AKT activation, which then regulates the activities of mTOR and FOXO1. The androgen-AR complex affects IGF-1 signaling via multiple ways, either in a transcription activity-dependent or transcription activity-independent manner. Dashed line indicates that the underlying mechanism is not fully elucidated.

FGF8 is another androgen-regulated growth factor (44). The tumorigenic effect of FGF8 has been demonstrated in both cell culture and transgenic mice (45, 46). FGF8 protein is overexpressed in human prostate cancers (47), and the level of expression correlates with tumor stage, pathological grade, and disease-specific survival (48). FGF8b protein expression is correlated with the AR expression in prostate cancer tissues and castration of mice significantly increases FGF8b expression in CWR22 prostate cancer xenografts, suggesting a role of androgen in the regulation of FGF8 expression. Indeed, androgens regulate FGF8 expression at the transcriptional level (49).

Prostate tumors also exhibit aberrant expression of EGF and EGF receptors (50–52). EGF signaling appears to be essential for the androgen-induced proliferation of LNCaP prostate cancer cells, since small molecule inhibitors against tyrosine kinase activity of the EGF receptor can completely suppress androgen-induced proliferation of these cells (52). Members of the EGF receptor family (ERBB1/EGFR, ERBB2, ERBB3, and ERBB4) may be differentially regulated by androgens. While androgens enhance the expression of EGFR, they reduce expression of ERBB2 in LNCaP cells (52, 53), suggesting that androgens regulate EGFR and ERBB2 via different mechanisms. The stimulation of EGFR gene expression by androgens appears to be at the transcriptional level, since androgen-induced EGFR upregulation does not require de novo protein synthesis. In contrast, androgen-induced reduction of ERBB2 does require de novo protein synthesis, suggesting that this repression is an indirect effect of androgens (53). Surprisingly, EGFR and ERBB2 expression in castration-recurrent 22Rv1 cells is not affected by androgen treatment. This discrepancy is most likely due to the presence of AR $\Delta$ CTD, a constitutively active form of AR, in 22Rv1 cells (53). AR $\Delta$ CTD is encoded by a splice variant mRNA of the AR gene, which has a novel exon 2b (54). Constitutively active AR variants promote expression of AR-regulated genes and cancer cell proliferation in the absence of androgens (54, 55). Taken together, these data suggest that AR regulates EGF/EGFR signaling during prostate cancer progression through either ligand-dependent or ligand-independent mechanisms.

Besides the regulation of growth factors and their receptors, androgens have also been shown to crosstalk with the downstream effectors of growth factor signaling, such as PI3K/AKT. The PI3K/AKT pathway is one of the most frequently altered signaling pathways in a variety of human cancers and plays a critical role in prostate carcinogenesis and its progression (56, 57). Constitutively activated AKT has been found frequently in prostate cancer cell lines and tissues, mostly due to loss of PTEN function (57). PTEN is a phosphatase which dephosphorylates PIP3, thereby inhibiting PI3K-induced AKT activation. Loss of

function of PTEN may be due to gene mutations (58), loss of heterozygosity (59), or gene deletions (60) in prostate cancers. Loss of function of PTEN and activation of AKT are significantly correlated with the progression of prostate cancer (61). Androgen-independent prostate cancer cell proliferation is correlated with increased activity of PI3K/AKT, suggesting a role of AKT in progression to castration-recurrent prostate cancers (CRPC) (62). Moreover, heterozygous  $PTEN^{+/-}$  mice develop low-grade PIN lesions in their prostates, while  $PTEN^{hy/-}$  mutants (harboring a hypomorphic allele with decreased PTEN expression) develop high-grade PIN lesions and locally invasive carcinomas (63). Moreover, loss of function of both PTEN and Nkx3.1, a well-known androgen-regulated transcription factor (64, 65), synergistically promotes prostate carcinogenesis (66).

Androgen-induced proliferation and survival of androgen-sensitive LNCaP cells depend on the activation of PI3K/AKT. Inhibition of AKT with a dominant-negative AKT or a PI3K inhibitor significantly attenuates androgen-induced cell proliferation (4, 5). Androgen-induced AKT activation requires the AR, but deletion of the ligand-binding domain of AR does not abolish it, indicating that this regulation is independent of AR transcriptional activity. Indeed, androgen-bound AR physically binds to the p85 $\alpha$  regulatory subunit of PI3K and thereby activates PI3K/AKT (4, 5). This is truly a two-way crosstalk, since AR expression and activity are also regulated by PI3K/AKT (67–69). Additional details about the crosstalk between androgen/AR and PI3K/AKT pathways have previously been discussed in several other reviews (61, 70).

The cell cycle, which is governed by the coordination of cyclins, cyclin-dependent kinases (CDKs) and CDK inhibitors, becomes deregulated in prostate cancer. Androgen/AR signaling is critical for normal prostate cell cycle progression, and dysregulation of this signaling may contribute to prostate cancer progression. Multiple mechanisms have been attributed to the effects of androgens on the cell cycle (71). For instance, androgens are important for cyclin D expression. Androgen-deprived prostate cancer cells arrest in early G1 phase, concomitant with loss of cyclin D, reduced CDK4 activity, and activated Rb. Addition of androgen rapidly increases cyclin D expression and promotes cell cycle progression (72, 73). Androgens increase cyclin D1 and D2 expression at the protein level but not at the mRNA level, suggesting that this regulation is at a post-transcriptional level. Androgen-induced cyclin D expression depends on activation of mammalian target of rapamycin (mTOR), which enhances the translation of cyclin D1 mRNA (73, 74). Although mTOR is a well-known target of AKT, androgen-induced mTOR activation is not mediated by AKT (73). The mechanism by which androgens activate mTOR remains unknown.

Androgens also regulate cell cycle progression through the CDK inhibitor p21<sup>Waf1/Cip1</sup>. Androgens positively regulate p21<sup>Waf1/Cip1</sup> expression through an ARE within its promoter (75). Induction of p21<sup>Waf1/Cip1</sup> by androgens may promote the assembly of active cyclin D1/CDK4 or CDK6 complexes (71, 76). In contrast to the upregulation of p21<sup>Waf1/Cip1</sup>, androgen treatment downregulates the expression of another CDK inhibitor p27<sup>Kip1</sup>, resulting in inhibition of cyclin E/CDK2 activity (77). This androgen-induced reduction of p27<sup>Kip1</sup> is mediated by decreased expression of the F-box protein SKP2 (78) which controls the ubiquitin-dependent degradation of p27<sup>Kip1</sup> (79, 80).

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#### 4. Androgen Action on Prostate Cell Apoptosis/ Survival

Androgens are essential for the survival of both normal and malignant prostate epithelium (21). Androgen withdrawal in adult rodents and humans induces apoptosis in the secretory epithelium (81, 82). Because most human prostate cancers are initially androgen responsive, androgen deprivation therapy remains the standard treatment for advanced prostate cancer. Recent data have revealed important pathways through which androgens regulate prostate cell apoptosis (83, 84).

Two types of signaling pathways lead to cell apoptosis: intrinsic or extrinsic. The intrinsic pathway is initiated by a variety of cell stresses, which lead to changes in mitochondrial permeability and release of cytochrome C and Smac/DIABLO. Antiapoptotic members of the BH3 family such as Bcl-2 and Bcl-XL inhibit intrinsic apoptosis by preventing the leakage of cytochrome C and Smac/DIABLO from mitochondria. Elevated Bcl-2 expression is implicated in a variety of human malignancies, including prostate cancer (85, 86). Whereas Bcl-2 expression is positive in most CRPCs, only approximately 30% androgen-dependent prostate cancers express low levels of Bcl-2, suggesting that Bcl-2 may contribute to the development of CRPC (87–89). It has been shown that androgens suppress Bcl-2 expression but the underlying mechanism is still unclear. Huang et al. reported that the regulation of Bcl-2 by androgens might be an indirect effect of AR activation, probably mediated by the E2F1 protein through a putative E2F-binding site in the promoter of the Bcl-2 gene (90).

Bak1 is another member of the BH3 family, which is also known to be regulated by androgens (91). Unlike Bcl-2, Bak1 is a proapoptotic protein. Under normal circumstances, Bak1 forms a complex with Bcl-2 or Mcl-1, which restrains Bak1 activation. Elimination of Bcl-2 or Mcl-1 caused by an apoptotic stimulus,

such as DNA damage, releases Bak1 from this inhibitory complex and promotes apoptosis (92). Bak-1 expression has been associated with prostate cancer progression. Bak1 is expressed in approximately 75% of primary and untreated localized prostate cancers, but in only approximately 33% of CRPCs (89). The suppression of Bak1 expression by androgens is mediated by a microRNA. MicroRNAs are a class of naturally occurring small RNAs, which do not encode proteins but regulate the expression of other genes. Androgens transcriptionally upregulate miR-125b, which then represses Bak1 expression (91). Thus, androgens suppress the expression of both Bcl-2 and Bak1 via different mechanisms. Further studies are essential to determine whether these androgen actions contribute to prostate carcinogenesis and cancer progression.

The extrinsic apoptotic pathway is mediated by death receptor signaling, which is triggered by ligands such as TNF- $\alpha$ , TRAIL, and FasL. Normal prostate cells are highly resistant to death receptor-induced cell apoptosis due to activation of NF- $\kappa$ B, which promotes cell proliferation and inhibits apoptosis (93). It has been reported that androgens inhibit TNF- $\alpha$ -induced NF- $\kappa$ B activation *via* multiple mechanisms (94–96) (Fig. 2.2). Keller et al. showed that androgens prevent the degradation of I $\kappa$ B $\alpha$ , which

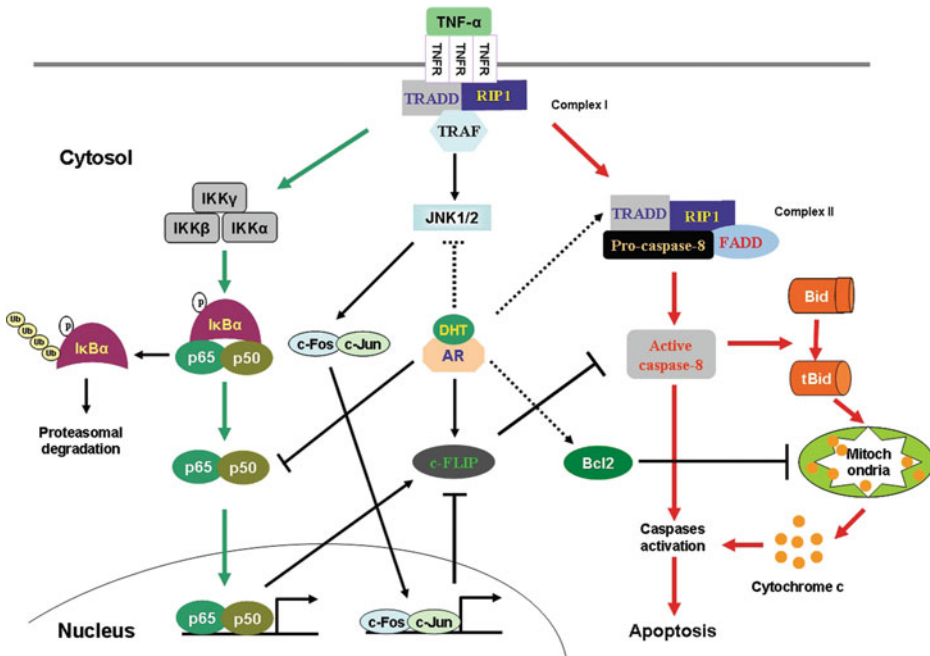


Fig. 2.2. Androgen action on TNF- $\alpha$ -induced signaling pathways. TNF- $\alpha$  binding to trimeric TNFR1 leads to assembly of complex I, which then results in activation of NF- $\kappa$ B, JNK, and/or the caspase cascade, depending on cellular context. All of these three TNF- $\alpha$  downstream signaling pathways can be regulated by androgen-AR via different mechanisms, as summarized in the text. *Dashed line* indicates that the underlying mechanism is not fully elucidated.



binds to NF- $\kappa$ B and inhibits its activation (95). AR activation also results in a decrease in RelA/p65, a subunit of NF- $\kappa$ B, thereby reducing its nuclear localization and transcriptional activity (97). Another mechanism whereby androgens affect NF- $\kappa$ B activity is through the formation of an AR-p65 complex via CREB-binding protein (CBP), which inhibits both p65 and AR transcriptional activities (94).

C-FLIP is a caspase-8 homologue, which functions as a dominant inhibitor of caspase-8 (98), thereby negatively regulating death receptor-induced apoptosis. In prostate cancer cells, increased c-FLIP expression is associated with increased resistance to death receptor-induced apoptosis (99, 100). LNCaP xenografts overexpressing c-FLIP are also resistant to castration-induced growth inhibition, suggesting a role of c-FLIP in the development of CRPC (99). It has also been observed that both c-FLIP mRNA and protein levels are reduced during progression to CRPC in animal models (101, 102). C-FLIP is directly regulated by androgens via a cluster of four AREs within a 156-bp region downstream from the transcription start site (99). Accordingly, both c-FLIP mRNA and protein levels are reduced following castration of rats in multiple tissues, including dorsolateral prostate and seminal vesicles (100). Unexpectedly, it has also been reported that androgen treatment downregulates c-FLIP in LNCaP cells, in which AKT is constitutively activated due to loss of PTEN. This discrepancy might be explained by the involvement of an AKT-regulated transcription factor, FOXO3a. Androgen induction of c-FLIP requires the presence of FOXO3a, which binds to the AR and potentially to the Forkhead-binding site within the c-FLIP promoter (102). Expression of FOXO3a TM, a constitutively active form of FOXO3a, rescues the androgen induction of c-FLIP in LNCaP cells, supporting a critical role of FOXO3a in androgen-induced c-FLIP upregulation (102).

FOXO3a belongs to the Forkhead transcription factor class-O family, members of which can act as tumor suppressors in a variety of malignancies (103, 104). Other members of the FOXO family include FOXO1, FOXO4, and FOXO6. AKT is a major regulator of the FOXO proteins. AKT phosphorylates the FOXO proteins, leading to their retention in the cytoplasm and proteasome-mediated degradation (105, 106) (Fig. 2.1). FOXO proteins inhibit cell proliferation and induce apoptosis in prostate cells. Thus, their activities are hypothesized to be reduced during prostate cancer progression (107, 108). Androgens negatively regulate the proapoptotic effects of FOXO1 through a physical interaction with it. Liganded AR blocks the DNA-binding activity of FOXO1 and impairs the ability of FOXO1 to induce Fas ligand expression and prostate cancer cell apoptosis (109). Moreover, androgen treatment results in a reduction of FOXO1 expression at the protein level via a proteolytic mechanism.

Treatment of LNCaP cells with androgens leads to FOXO1 protein cleavage and produces a truncated FOXO1, which lacks ~120 amino acid residues in the C-terminus. Ectopic expression of this truncated FOXO1 inhibits the transcriptional activity of the intact FOXO1, suggesting that androgen-induced FOXO1 protein cleavage results in reduction of FOXO1 transcriptional activity (107).

TRADD (TNF receptor-associated death domain), which is a transducer for death receptor-induced signaling, is also a target of androgens (110). TRADD mediates TNFR1-induced apoptosis as well as NF- $\kappa$ B activation (111). Overexpression of TRADD in a variety of cell lines leads to apoptosis; however, knockdown or knockout of TRADD expression in some cell lines does not inhibit apoptosis, suggesting that its function depends on the cellular context (112–114). TRADD protein is reduced in CRPC cells compared to androgen-responsive cells. Androgen deprivation reduces TRADD expression in prostate cancer cell lines, xenografts, and human tissues. Moreover, androgen treatment increases TRADD expression at both the mRNA and protein levels (110). Unpublished data (D. Wang, personal communication) suggest that this regulation is an indirect action of androgens because it requires AR and de novo protein synthesis. Reduced TRADD expression may account for the reduced sensitivity of CRPC cells to TNF- $\alpha$ .

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## 5. Androgen Action on Prostate Cancer Metastasis

A pivotal problem of prostate cancer, as in other cancers, is its propensity to metastasize. The process of tumor metastasis includes the following: activation of epithelial–mesenchymal transition (EMT), remodeling of the extracellular matrix, neovascularization, and migration to specific secondary sites. Many of the androgen-regulated signaling pathways that were discussed above are also important for prostate cancer metastasis. For instance, NF- $\kappa$ B activity has been associated with many types of metastases. Inhibition of NF- $\kappa$ B activity in metastatic prostate cancer is associated with reduced expression of vascular endothelial growth factor, interleukin-8, and matrix metalloproteinase-9 and a concomitant decrease in angiogenesis, invasion, and metastasis in nude mice (115). Furthermore, nuclear NF- $\kappa$ B expression in primary prostate cancers is highly predictive for pelvic lymph node metastases (116). Thus, the regulation of prostate cancer metastasis by androgens may be achieved by crosstalk between androgen and NF- $\kappa$ B signaling pathways.

Cell adhesion molecules such as cadherins play a critical role in the activation of EMT (117). Cadherins are a class of type-1 transmembrane glycoproteins, of which E-cadherin and N-cadherin are the best characterized in prostate cancers. E-cadherin is an important tumor suppressor gene. Loss of E-cadherin expression disrupts cell–cell junctions and consequently promotes cell migration, leading to tumor metastasis. Multiple studies reported that loss of E-cadherin expression enhances the progression from nonmetastatic to metastatic carcinoma (118, 119). In primary prostate cancers, reduced E-cadherin expression has been correlated with increased tumor grade, bone metastasis, and poor prognosis (120, 121). In contrast, increased levels of N-cadherin and cadherin-11 are associated with poorly differentiated and metastatic prostate cancers (122, 123). In more aggressive prostate cancer specimens, N-cadherin expression is increased while E-cadherin is reduced. This phenomenon is called ‘cadherin switching’ (124). Interestingly, both E-cadherin and N-cadherin appear to be regulated by androgens (125, 126). Androgen deprivation therapy leads to elevated expression of E-cadherin in prostate cancer, suggesting that androgens repress E-cadherin expression (127). Moreover, androgen treatment reduces E-cadherin expression in the breast cancer cell lines MCF7 and T47D. Identification of AREs within the promoter of the E-cadherin gene further suggests that this repression is a direct transcriptional effect of AR (125). However, because these studies were not conducted in prostate cancer cell lines or models, it is still not clear whether similar mechanisms apply to prostate cancer cells. More studies are needed to elucidate the role of androgens on E-cadherin expression and function in prostate cancer.

In contrast to E-cadherin, N-cadherin is induced by androgen deprivation in experimental castration-recurrent prostate cancer models as well as in human prostate tumors (126). It has also been shown that testosterone increases N-cadherin expression in motor neurons (128). Taken together, these results suggest that androgens may positively regulate the expression of N-cadherin.

Cadherin-11 is a homophilic cell adhesion molecule that mediates osteoblast adhesion and thereby plays a critical role in the metastasis of prostate cancer to bone. Increased cadherin-11 expression correlates with development of CRPC (129). Cadherin-11 appears to be indirectly regulated by androgens, suggesting a role of androgen in the metastasis of prostate cancer to bone (130).

Maspin is a serine protease inhibitor with tumor suppressing properties. Loss of maspin is associated with a variety of tumors, such as breast and prostate cancers (131, 132). Consistently, re-expression of maspin in prostate cancer cells inhibits tumor growth and prostate cancer-induced bone remodeling (133).

Maspin exerts its anti-metastasis activities via multiple mechanisms. For instance, maspin blocks FGF and vascular endothelial growth factor-mediated endothelial cell migration *in vitro*. Maspin also inhibits prostate cancer growth and tumor angiogenesis *in vivo*, probably mediated by its ability to inhibit the degradation of extracellular matrix via tPA and pro-uPA (134–136). Maspin expression increases in prostate cancer cells and tissues following androgen deprivation while androgen treatment decreases maspin expression (132, 137). Identification of an ARE element in the promoter of the maspin gene has confirmed this regulation as a direct transcriptional effect of the androgen receptor (137). Therefore, androgens may affect prostate cancer progression and metastasis via regulation of maspin expression.

**Table 2.1**  
**Androgen-regulated factors associated with prostate carcinogenesis**

Target	Function	Mechanism	References
TMPRSS2-ETS	Transcription factors	Direct transcriptional regulation	(24, 29)
IGF-1	Growth factor	Direct transcriptional regulation	(38)
IGF-1 R	Growth factor receptor	Nongenomic effect	(39)
FGF8	Growth factor	Direct transcriptional regulation	(49)
EGFR	Growth factor reception	Direct transcriptional regulation	(53)
ERBB2	Growth factor receptor	Indirect transcriptional regulation	(53)
AKT	Protein kinase	Nongenomic effect	(4, 5)
Cyclin D	Cell cycle regulator	Indirect regulation mediated by mTOR	(73)
P21	CDK inhibitor	Direct transcriptional regulation	(75)
P27	CDK inhibitor	Indirect regulation mediated by SKP2	(78)
Bcl-2	Anti-apoptotic protein	Indirect transcriptional regulation	(90)
Bak1	Pro-apoptotic protein	Indirect transcriptional regulation mediated by miR-125b	(91)
C-FLIP	Anti-apoptotic protein	Direct transcriptional regulation	(99)
FOXO1	Transcription factor	Indirect effects on protein degradation	(107)
TRADD	Signaling transducer	Indirect transcriptional regulation <sup>a</sup>	(110)
E-cadherin	Cell adhesion molecule	Direct transcriptional regulation	(125)
Cadherin-11	Cell adhesion molecule	Indirect transcriptional regulation	(130)
Maspin	Serine protease inhibitor	Direct transcriptional regulation	(137)

<sup>a</sup>Personal communication with D. Wang

## 6. Conclusions

It has long been recognized that androgens play a critical role in prostate carcinogenesis. In this review we have highlighted several androgen-regulated signaling pathways and factors that may be involved in prostate carcinogenesis (**Table 2.1**). Although under certain circumstances androgens may inhibit cell proliferation or promote cell death, in general, it is well accepted that androgens are critical for prostate cancer cell proliferation and survival. However, the effects of androgens are diverse and complex, and focusing on one or two signaling pathways for delineating mechanisms is likely to be an oversimplification. More importantly, the downstream signaling pathways of androgen may also crosstalk with each other, making the contributions of androgen to prostate carcinogenesis more complicated. Nonetheless, recent development of more potent antiandrogens and inhibitors of androgen metabolism, which are in clinical trials (**138–140**), will aid in understanding the role of androgen signaling in prostate cancer cells. Taken together, better understanding of AR action is likely to lead to better clinical treatments for prostate cancer.

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