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Review

Recent advances in androgen receptor action

H.-J. Lee[a] and C. Chang[b,*]

[a] Institute of Biotechnology and Department of Life Science, National Dong Hwa University, Hualien 974 (Taiwan)
[b] George H. Whipple Laboratory for Cancer Research, Departments of Pathology, Urology, Radiation Oncology, and the Cancer Center, University of Rochester, Rochester, New York 14642 (USA), Fax: +1 585 756 4133, e-mail: chang@urmc.rochester.edu

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Abstract. Androgens, principally testosterone and 5α-dihydrotestosterone, play critical roles in the development and growth of the male reproductive and nonreproductive systems. Their biological actions are mediated by the androgen receptor (AR), a ligand-dependent transcription factor, belonging to the nuclear receptor superfamily. These androgen-AR complexes interact with various factors (e.g. coactivators or corepressors) to modulate transcription of androgen target genes via specific DNA sequences. Many lines of evidence have also correlated AR with several mammalian disorders. Finally, recent advances in molecular biology have significantly impacted our knowledge of the role of AR in mammals. The aim of this review is to present recent emerging aspects of AR action.

Key words. Androgen; coactivator; nuclear receptor.

Introduction

Androgens mediate a wide range of developmental and physiological responses, and are especially important in the male reproductive and nonreproductive systems [1, 2]. The effects of androgens are mediated through the androgen receptor (AR), a member of the nuclear receptor (NR) superfamily [3]. NR members form the largest known superfamily of genes encoding ligand-inducible transcription factors that regulate complex gene networks in a wide variety of biological processes, such as growth, development, and differentiation [4]. Upon binding to their respective ligands, NRs may undergo a transformation step, bind to a specific DNA sequence called a hormone response element (HRE) and then regulate the transcription of their target genes [5].

* Corresponding author.

Androgen receptor functional domains

The human AR complementary DNA (cDNA) was initially cloned in 1988 by Chang et al. [6, 7], Lubhan et al. [8] and others soon thereafter [9–11]. AR is a 110-kDa nuclear protein that contains 918 amino acid residues. Throughout the text, the numbering system of AR codons will be based on a primary sequence of 918 amino acids [6, 7]. The androgen-AR complex can either induce or suppress the androgen-responsive genes via binding to a specific HRE, known as an androgen response element (ARE) [1, 12]. As shown in figure 1, AR consists of separate domains responsible for ligand binding (LBD), DNA binding (DBD), nuclear localization, and transcriptional modulation [3, 4]. The AR gene containing eight exons is located on the proximal long arm of the X chromosome at Xq11–12 [8, 11]. The well-conserved DBD is a 68-amino acid region which can fold into two zinc-coordinated finger structures with the ability to bind to DNA. The 295-amino acid region behind the DBD, including the hinge region and

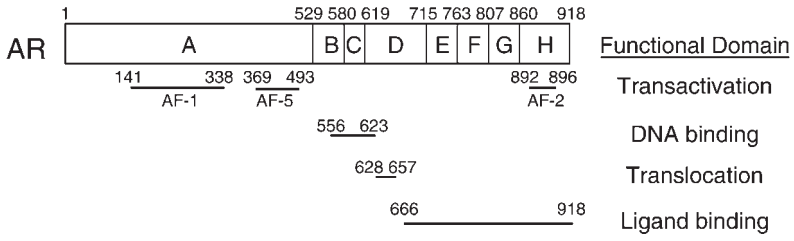

Figure 1. Schematic illustration of the functional domains of the human AR. The exons are labeled A–H. The numbers above the domains indicate the amino acid residue number at the junctions between exons and introns. Domains responsible for specific functions are also indicated by lines with the appropriate amino acid residue numbers.

the LBD, is responsible for dimerization and androgen binding [13]. Moreover, a nuclear localization signal (NLS) consisting of several clusters of basic amino acids is located within the second zinc finger and the hinge region. This NLS is responsible for nuclear trafficking of AR [14].

The most variable region is the N-terminal domain (NTD) with 555 amino acid residues. Both structural and functional studies have revealed that the NTD of AR contains one region referred to as AF-1, ranging from amino acids 141 to 338, which could be involved in transcriptional activation [15]. Recently, a second transactivation domain called AF-2 was identified in the LBD [16, 17]. This weak activation domain contains five conserved amino acids as its core region, although the boundaries of this domain have not yet been determined. A third transactivation domain referred to as AF-5 has been mapped from amino acids 369 to 493 in the NTD [17]. A possible interaction between the NTD and the LBD is highly speculated, and some regions of the NTD (but not AF-1) interact with AF-2 via a protein surface [17].

Phosphorylation of androgen receptor

Several reports have suggested that phosphorylation may influence NR transactivation, and in the case of AR, phosphorylation may result from rapid posttranslational modification within 10 min of AR synthesis (i.e. an androgen-independent manner), androgen binding, or association with several cellular protein kinases [1, 17–19].

In LNCaP prostate cancer cells, phosphorylation of AR can be demonstrated in the wild-type AR and in a truncated AR lacking the LBD. However, in this same series of experiments, no phosphorylation could be detected in a mutant AR without the NTD, indicating that most of the androgen-dependent phosphorylation sites on AR may be located in the NTD [1]. This NTD phosphorylation has been associated with the transcriptional regulatory properties of the receptor.

In summary, phosphorylation of AR may cause some conformational changes that modify the ligand and DNA-binding properties of AR. It has been proposed that there are two phosphorylation steps involved in AR activation, the first one being a rapid posttranslational modification for the acquisition of the androgen binding. The second phosphorylation step then occurs upon hormone binding [17]. Recently, both Akt and Mdm2 have been shown to form a complex with AR, and promote phosphorylation-dependent AR ubiquitylation, resulting in AR degradation by the proteasome [20]. More studies are needed to determine the precise role of phosphorylation in AR function.

Androgen target genes

AR regulates the expression of androgen-responsive genes by binding to AREs located mainly within the promoter region of target genes. Molecular cloning and sequence analysis of androgen-responsive genes have led to the identification and characterization of many AREs [1]. AR binds as a dimer to AREs that can be divided into two main categories: the inverted and direct repeat AREs [21]. The inverted repeat ARE, GGTACAnnnTGTTCT, is found in the mouse mammary tumor virus and prostate-specific antigen promoters, and is able to bind and mediate transcriptional activation by glucocorticoid receptor (GR) in some promoter contexts [22]. AR specificity for this category of response element may be determined by the relative abundance of the two receptors in a particular cell type, chromatin conformation, or ability to interact with transcription factors flanking the response element. The first zinc finger of AR recognizes and makes direct contact with the inverted repeat ARE, while the second zinc finger of AR stabilizes the receptor-DNA interaction by contacting the DNA phosphate backbone [21]. In contrast, the direct repeat ARE is found in a number of androgen response genes, including probasin and sex-limited protein. The direct repeat ARE is specific for AR, but not for GR [23]. Recognition of the direct repeat ARE is apparently mediated by the second zinc finger and part of the hinge region of AR [21]. Polymorphic variants of AR have been shown to have reporter gene-specific tran-

scriptional effects, suggesting that different AREs may also influence the conformation and protein-protein interaction ability of AR.

Molecular mechanisms of AR action

Coregulators

Androgens [testosterone (T) and 5α-dihydrotestosterone (DHT)] freely diffuse into all cells (fig. 2). The actions of androgens are mediated by AR, which normally binds T or DHT with high affinity (0.1 nM) and limited capacity. In the presence of ligand, AR undergoes a change in conformation, resulting in an activated form that interacts tightly as a homodimer with a specific ARE. The hormone-receptor complex then activates transcription of target genes. This in turn results in protein translation, and subsequent alterations in cell function. Recent advances in molecular biology have led to the identification of many coregulators (coactivators or corepressors) that interact with receptor proteins via protein-protein interactions, and play important roles in transcriptional activation (table 1) [5, 24, 25].

Coactivators

Coactivators are proteins that enhance ligand-dependent transcriptional activity. They may serve as bridging or adaptor molecules, binding to NRs themselves, recruiting additional proteins [e.g. histone acetyltransferases (HATs)], and interacting with the basal transcriptional machinery to enhance transcription of target genes. Several biochemical and molecular screening approaches, such as the yeast two-hybrid system and Far-Western blotting, have led to identification of a large number of potential coactivator proteins. Most of the AR coactivators identified to date also stimulate the activity of other NRs and transcription factors [26]. The list of AR coactivators has been growing rapidly [26-29], and some important AR coactivators are summarized in the Appendix. The relative importance of each coactivator and its precise functions, however, are still unknown.

Corepressors

Recent studies utilizing the yeast two-hybrid system and other techniques have identified corepressors that function as negative regulators to repress or silence basal transcriptional activity [5]. Some important AR corepressors are summarized in the Appendix.

Cross-talk with nuclear receptors or other systems

Androgenic regulation of male reproductive system development and maintenance, as well as prostate cancer progression, has traditionally been viewed in terms of AR transactivation. With the identification of numerous AR coregulators, however, it is likely that many other signaling pathways influence androgen action via crosstalk with AR.

As previously mentioned, ARA70 [30], TR4 [64], SRC family members [50-53], and CBP/p300 and their associated proteins [42, 43] are AR coregulators. Through interaction with coregulators, such as BRCA1 [39] and Smad3 [46], and components of different pathways, such as akt [19] and oncoproteins [18, 42], AR may be involved in apoptosis, (TGFβ), mitogen-activated protein (MAP) kinase and activator protein-1 (AP-1) signaling

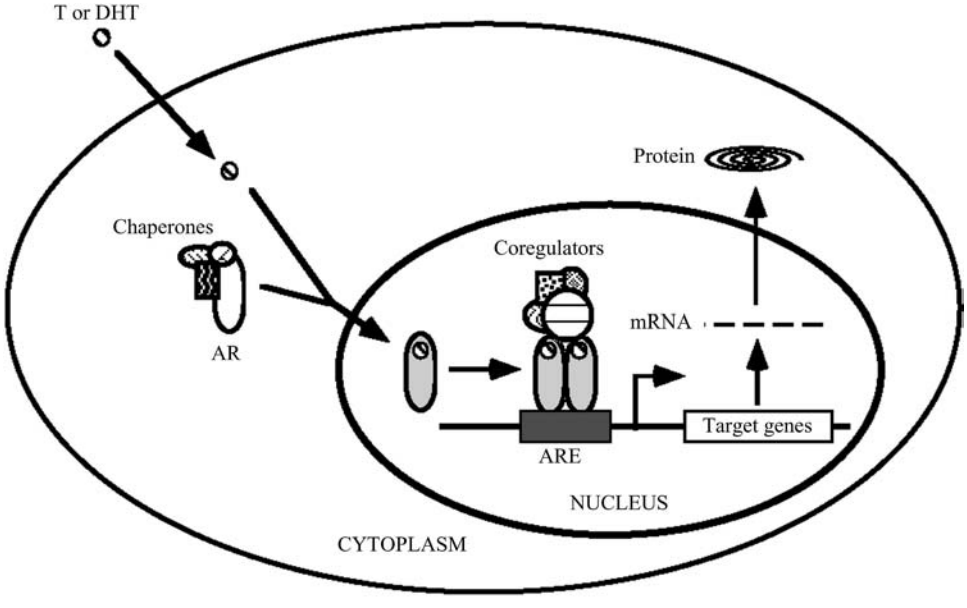

Figure 2. Molecular mechanism of AR action.

Table 1. AR coregulators.

Function	Name	Molecular mass (kDA)	Interacting domain of AR	Reference
Coactivator	ARA70 (RFG, ELEIα, NCOA4)	70	DBD-LBD[a]	30
	ARA55 (Hic-5)	55	LBD	34
	ARA54	54	LBD	35
	ARA24 (Ran)	24	NTD	36
	ARA160 (TMF)	160	NTD	37
	ARA267 (NSD1)	267	NTD, LBD	38
	BRCA1	190	NTD, LBD	39
	CAK (TFIIH)	39	NTD	40
	CBP	300	NTD, LBD	41
	P/CAF	62	LBD	43
	P-TEFβ	81	NTD, DBD	44
	Rb	110	NTD, DBD	45
	Smad3	150	DBD, LBD	46
	SRA	–[b]	–[c]	47
	SRC-1 (NcoA-1)	160	NTD, LBD	50
	Supervillin	205	NTD, DBD-LBD	54
	TIF2 (GRIP1)	160	LBD	51, 52
	Tip60	60	LBD	55
	TRAM-1	155	–[c]	53
Corepressor	Cyclin D1	33	NTD	56
	HBO1	71	DBD, LBD	57
	p53	53	NTD-LBD	58
	RelA	65	NTD	59
	PIASy	57	DBD	60
	PyK2	116	LBD-ARA55[d]	61
	TR4	67	DBD, LBD	64

[a] DBD, LBD and NTD represent interacting area of the DNA-binding, ligand-binding, and N-terminal domains of AR, respectively.
[b] SRA is an RNA, 883 nucleotides in length.
[c] Not determined.
[d] PyK2 interacts with ARA55, which binds to the ligand-binding domain of AR.

pathways. Further characterization of the cross-talk between AR and other cell signaling pathways may provide novel insight into the development of more effective therapeutic agents for prostate cancer.

Chaperones

Molecular chaperones assist proteins in assuming a mature and functional conformation [65]. It has become apparent in recent years that chaperones function as part of a multiprotein heterocomplex that is potentially involved not only in protein folding, but also in intracellular trafficking and in targeting proteins for degradation. In the case of NRs, the activity of the chaperone heterocomplex, as well as the proteins comprising the heterocomplex, has an effect on the observed ligand-dependent transcriptional activity of the receptor. The direct interaction between chaperones and NRs makes them potential therapeutic targets in a number of pathologic conditions. In the case of cancers involving NRs, such as breast and prostate cancer, the inhibition of chaperone activity may inhibit tumor cell growth. Conversely, enhancement of chaperone activity may be beneficial in disorders involving protein misfolding, as in the case of AR aggregates found in Kennedy's disease.

AR and disease

Androgen insensitivity syndrome (testicular feminization)

Mutations in the AR gene are thought to cause androgen insensitivity syndrome (AIS) in subjects with the 46,XY karyotype [1]. AIS may be the most commonly described hormone insensitivity syndrome. One of the major clinical manifestations of AIS is testicular feminization (Tfm) syndrome, where male sexual development is deterred or absent.

We have studied tissues from several cases of AIS-affected individuals in our laboratory [66–71], and detailed reports were reviewed [1]. The AR gene mutations database has the most recent information, including over 500 reported mutations in the July 2001 version [72]. The database is available on the Internet at http://www. mcgill.ca/androgendb/ [EMBL-European Bioinformatics Institute (ftp.ebi.ac.uk/pub/databases/androgen), or as a Macintosh File-

makerPro or Word file (MC33@musica.mcgill.ca)]. In summary, mutations of the AR gene are associated with a variety of diseases. However, the majority of mutations identified to date are linked to AIS. These mutations can result in a truncated receptor or altered affinity for ligand. Whatever the nature of the mutation, the end result is diminished AR function. Future studies will uncover further associations between AR mutation and disease.

Prostate cancer

Tumors forming in androgen responsive tissues, such as the prostate, frequently express AR, and androgen ablation therapy is the main treatment for advanced prostate cancer [73]. Despite the initial response to this therapy, prostate cancer invariably progresses to an androgen-independent state [74]. The molecular mechanisms underlying this progression may be potential targets to manage advanced prostate cancer growth. The link between AR mutations and prostate cancer progression is controversial, since the frequency of AR mutations in primary prostate cancer is low [26, 75]. Conversely, studies have shown a much higher incidence of AR mutations in metastatic prostate cancer [75]. These results suggest that as prostate tumors metastasize to local lymph nodes, the frequency of AR mutations rises. Amplification of the AR gene is another potential mechanism of androgen-independent growth and is detected in 30% of prostate tumors. Transcriptional activation of AR resulting from cross-talk with other signal transduction pathways, the proteasome system, and/or interaction with AR coregulators, may also cause receptor activation and cell proliferation in prostate cancer [29, 76, 77].

Skin and hair follicles

AR is expressed in the skin (e.g. hair follicles, sebaceous glands and apocrine glands) and in genital skin fibroblasts [26]. Moreover, acne is a classical androgen-mediated disease.

Androgens regulate many aspects of human hair growth in both sexes [17]. Hirsutism, or increased facial hair in women, is frequently due to elevated circulating androgen levels [26]. The actions of androgens on hair in men are location dependent. Androgens suppress hair growth in male pattern baldness.

Among androgen targets in the skin, hair follicles have been most intensively studied in terms of androgen function [78, 79]. Hair follicles consist of mesenchymal cells (e.g. dermal papilla cells and connective root sheath cells) and epithelial cells (e.g. outer root sheath cells, inner root sheath cells and matrix cells). According to the current hypothesis, androgens control most follicular cells indirectly via the mesenchyme-derived dermal papilla which regulate many aspects of follicular activity [80]. In this model, androgens bind to AR in dermal papilla cells, altering their production of unknown regulators that influence other follicular components. These unknown regulators may be soluble paracrine factors and/or extracellular matrix proteins. Identification of unknown regulators should lead to a greater understanding of androgen action in hair follicles, enabling the development of better treatments for associated androgen-potentiated disorders [80].

Immune function

Androgens have been shown to modulate the hematopoietic and immune systems, and have been used clinically to stimulate hematopoiesis in bone marrow failure conditions [26, 81, 82]. Evidence shows that AR is ubiquitously expressed in the bone marrow in both sexes, but is not detected in lymphoid or erythroid cells, or eosinophils [81]. In addition, the AR expression pattern does not change with age, indicating that androgens may exert direct modulating effects on a wide spectrum of bone marrow cell types via AR-mediated responses.

Kennedy's disease (spinal and bulbar muscular atrophy)

Mutations in AR are also associated with spinal and bulbar muscular atrophy (SBMA), or Kennedy's disease, an X-linked recessive genetic disorder characterized by progressive muscular weakness, cramps and twitching in the limbs in adults [83]. Affected males have testicular atrophy, reduced fertility, and excessive development of the male mammary glands (gynecomastia). The AR of these patients has an increased number of polymorphic tandem CAG repeats in the coding region of exon 1 [84]. This triplet CAG, located in exon 1 of the AR gene and encoding a polyglutamine stretch, is amplified in patients to about twice the normal size (38–75 compared with 9–33) [85]. Interestingly, a somatic mutation of the AR CAG repeat has also been detected in a patient with prostate cancer [86], with tumor DNA containing one PCR fragment with 24 CAGs (wild-type) and a second fragment with 18 CAGs (mutant).

The presence of a CAG repeat in AR may influence its transcriptional activity and in vivo function. Nonetheless, the role of this AR polymorphism in Kennedy's disease is still unclear. Evidence has been presented suggesting that aggregate formation and proteolytic processing of AR protein can occur in a polyglutamine repeat length-dependent manner, and aberrant metabolism of the expanded repeat AR is linked to cellular toxicity [85].

More recently, caspase-3 cleavage of an AR with an expanded polyglutamine tract was shown to play a role in the induction of neural cell death [85]. Interestingly, ARA24 can bind differentially to different lengths of

polyQ within AR. The reduced interaction and weaker coactivation of the longer polyQ AR by ARA24 may contribute to partial androgen insensitivity during the development of Kennedy's disease [36].

Conclusions

The continuing study of AR coregulators has revealed potentially multiple mechanisms through which the transcription activity of AR may be modulated [21, 29, 87]. However, the mechanism of action and relative in vivo importance of AR coregulators have not been established yet. Coregulators are typically identified on the basis of interaction studies, and their influence is evaluated by transient transfection. Thus far, questions of their role in development, or in pathological or physiological conditions, remain unanswered.

Androgens are thought to predominantly mediate their biological effects through binding to AR. Interestingly, more evidence has indicated that androgens, like progesterone and estrogen, can exert nongenomic effects [88]. Nongenomic steroid activity typically involves the rapid induction of secondary messengers of signal transduction cascades, including increases in free intracellular calcium, and activation of protein kinase A (PKA), protein kinase C (PKC), and MAP kinase [89]. Such nongenomic androgen effects occur in cell types that lack a functional AR, in the presence of inhibitors of transcription and translation, or are observed to occur too rapidly to involve changes in gene transcription. The physiological effect of nongenomic androgen action has yet to be determined. However, it may ultimately exert a biological effect through modulation of the transcription activity of AR or other transcription factors. In addition, such modulation may occur through direct phosphorylation of transcription factors or their coregulators.

Recently, AR knockout mice have been generated and characterized [90]. Phenotype analyses showed that AR knockout male mice have a female-like appearance and body weight. Their testes are 80% smaller, and serum T concentrations are lower than those of wild-type mice. Spermatogenesis is arrested in pachytene spermatocytes. The number and size of adipocytes are also different between the wild-type and AR knockout mice. Cancellous bone volumes of AR knockout male mice are reduced compared with wild-type littermates. In addition, the average number of pups per litter in homologous and heterozygous AR knockout female mice is lower than that in wild-type female mice, suggesting potential defects in female fertility and/or ovulation.

In summary, AR plays an important role in development and differentiation in health and disease. Advances in molecular biology have significantly impacted our knowledge of the role of this important member of the NR superfamily. We have just begun to elucidate the complexity of AR action. Subsequently, additional effort is required to fully understand the detailed mechanism of AR action.

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Appendix

1. Coactivators

ARA70. Using a yeast two-hybrid system with a Gal4-AR fusion protein as bait to screen a human brain cDNA library, ARA70 was isolated as the first ligand-dependent AR coactivator [30]. ARA70 (RFG, ELE1α, NCOA4) enhances the transcriptional activity of AR in prostate cancer DU145 cells [26-32]. In addition to AR, ARA70 is able to interact with the peroxisome proliferator-activated receptor γ (PPARγ) [33].

ARA55. Isolated by a yeast two-hybrid system, ARA55 consists of 444 amino acid residues with a predicted molecular mass of 55 kDa [34]. The sequence of ARA55 shows very high homology to mouse hic-5, a TGFβ-inducible gene. The C-terminal half of ARA55 contains three LIM motifs and is sufficient for interaction with AR.

ARA54. ARA54 is another AR coactivator isolated using a yeast two-hybrid system [35]. ARA54 consists of 474 amino acids with a predicted molecular mass of 54 kDa. Interestingly, ARA54 can enhance the transcriptional activity of the LNCaP mutant AR (T877A), but not the wild-type AR or another mutant AR (E708K) in response to 17β-estradiol or hydroxyflutamide.

ARA24. A nuclear G protein, Ras-related nuclear protein/ARA24 was isolated as the first AR coactivator that can bind differentially with different lengths of polyglutamine (polyQ) within AR [36]. Poor interaction and weaker coactivation of the longer polyQ AR from Kennedy's disease by ARA24 may contribute to the weaker transactivation of this AR mutant.

ARA160. ARA160, containing 1093 amino acids with an apparent molecular mass of 160 kDa, was isolated by Far-Western blotting [37]. ARA160 is an AR N-terminal coactivator, and its sequence is identical to that of the HIV-1 TATA element modulatory factor (TMF). Furthermore, ARA160 can enhance AR transactivation cooperatively with ARA70 in PC-3 prostate cancer cells.

ARA267. Recently, ARA267, an exceptionally large protein with 2427 amino acids and a predicted molecular mass of 267 kDa, was isolated [38]. ARA267 contains one SET domain, two LXXLL motifs, three NLSs, and four plant homodomain finger domains (PHDs). In addition, ARA267 can cooperate with other coactivators, such as ARA24 and P/CAF, to enhance AR transactivation.

BRCA1. Breast cancer susceptibility gene 1 (BRCA1) functions as a tumor suppressor, and regulates other cellular processes [39]. Recently, BRCA1 was linked to the suppression of ER transactivation. BRCA1 interacts with AR and enhances AR target gene expression, such as p21(WAF1/CIP1), potentially resulting in the increase of androgen-induced cell death in PC-3 cells. In addition, BRCA1 and other AR coregulators, such as ARA55 and ARA70, can synergistically enhance AR transactivation.

CAK. AR interacts with the general transcription factor TFIIH under physiological conditions. Cdk-activating kinase (CAK), the kinase moiety of TFIIH, is an AR coactivator [40]. These results suggest that AR may interact with TFIIH for efficient communication with the general transcription factor/RNA polymerase II complex on the core promoter of target genes.

CBP and its associated proteins. CREB-binding protein CBP (mouse)/p300 (human) are large nuclear proteins that can interact with a variety of different DNA-binding proteins [5]. These proteins are classified as cointegrators, but are sometimes also considered coactivators. Cointegrators are common limiting cofactors, which may synergistically coordinate the transcriptional effects of NRs with the basal transcription machinery. CBP is an AR coactivator [41, 42], and the 12S E1A adenoviral protein that inactivates CBP function also inhibits AR transactivation. In addition, the CBP/p300-associated factor (P/CAF), which possesses HAT activity, is also an AR coactivator [43].

P-TEFb. A kinase subunit of positive elongation factor b (P-TEFb) interacts with AR, and enhances the efficiency of AR transactivation in PC-3 cells [44]. A nuclear run-on transcription assay suggested that this enhancement is involved in the elongation stage of transcription.

Rb. Using a glutathione S-transferase (GST) pull-down assay, the retinoblastoma (Rb) protein was identified as an AR coactivator in human prostate cancer DU145 cells [45]. Rb has a well-characterized role in cell growth inhibition by sequestering transcription factors. In the case of AR, however, Rb plays a positive role by enhancing AR transcriptional activation.

Smad3. A downstream mediator of the TGFβ signaling pathway, Smad3 also functions as an AR coactivator [46]. Endogenous prostate-specific antigen (PSA) expression in LNCaP cells can be induced by DHT, and expression of the Smad3 further induces PSA expression.

SRA. Steroid receptor RNA activator (SRA) functions as an RNA coactivator of several NRs, including AR [47]. SRA may confer functional specificity upon multiprotein complexes recruited by liganded receptors during transcriptional activation.

SRC members. The steroid receptor coactivator (SRC) family comprises a number of NR coactivators with a molecular mass of about 160 kDa [5]. This family contains three subgroups, SRC-1, SRC-2, and SRC-3 [48, 49]. The SRC-1 subgroup contains human SRC-1, and its mouse homolog, NCoA-1 [50]. The SRC-2 subgroup consists of TIF2 (mouse) and GRIP1/NCoA (human), while the SRC-3 subgroup has ACTR, AIB1, RAC3, TRAM-1 and xSRC-1. Evidence indicates that SRC-1 [50], TIF2 [51], GRIP1 [52], and TRAM-1 [53] are AR coactivators.

Supervillin. Supervillin (SV), an actin-binding protein, isolated from the skeletal muscle library, is an AR coregulator [54]. SV can cooperate with other AR coactivators, such as ARA55 or ARA70, to enhance AR transactivation. However, SV shows a mild suppressive effect on amino- and carboxyl-terminal interactions of AR, suggesting SV may go through a different mechanism to enhance AR transactivation.

Tip60. Tip60 was isolated as an AR coactivator by yeast two-hybrid screening [55]. It was originally identified as a coactivator for the HIV TAT protein. Tip60 can enhance AR-mediated transactivation in a ligand-dependent manner in LNCaP and COS-1 cell lines. Moreover, Tip60 enhances transactivation to levels observed with SRC-1 and CBP, confirming its significance as an AR coactivator.

Others. Many other AR coactivators have also been identified (see [29] for details). The number of AR coactivators identified is growing, suggesting many potential levels of cross-talk between androgen and other cell signaling pathways.

2. Corepressors

Cyclin D1. Cyclin D1 phosphorylates Rb, promoting cellular proliferation, and inhibiting cellular differentiation in several different cell types. Cyclin D1 is also an AR corepressor that represses ligand-dependent AR activity by directly binding to the N terminus of AR, as well as competing for P/CAF [43, 56].

HBO1. Histone acetyltransferase binding to ORC (HBO1) is a nuclear protein with the highest expression levels in human testis. HBO1, belonging to the MYST family, may specifically repress AR-mediated transcription in both CV-1 and PC-3 cells [57].

p53. The tumor suppressor protein p53 appears to be an AR corepressor by disrupting amino- and carboxyl-terminal interactions of AR [58]. This interaction is thought to be responsible for the in vivo homodimerization of AR. In addition, c-Jun is able to relieve the negative effects of p53 on AR transactivation.

RelA. RelA is an AR corepressor that interacts with AR and represses AR-mediated transactivation in a dose-dependent manner in COS-1 cells [59]. The repression of AR appears to involve the N-terminal region, between residue 296 and the DBD.

PIASy. The protein inhibitor of activated STAT (PIAS) family has two AR coactivators, PIAS1 and PIAS3 [60]. In contrast, PIASy is a transcriptional corepressor of AR.

This suggests that different PIAS proteins have distinct effects on AR signaling in prostate cancer cells.

Pyk2. The proline-rich tyrosine kinase 2 (Pyk2) is an ARA55-interacting protein that can repress AR transactivation via interaction of ARA55, an AR coactivator [61]. This inactivation may result from the direct phosphorylation at tyrosine 43 of ARA55 by Pyk2.

TR4. The testicular orphan receptor-4 (TR4) is a member of the NR superfamily [62, 63]. TR4 heterodimerizes with AR and represses AR target gene expression [64]. Interestingly, AR is also a TR4 corepressor. Simultaneous expression of both receptors may result in bidirectional suppression of their target genes.

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