Asian Journal of Andrology (2014) 16, 555–560

www.asiaandro.com; www.ajandrology.com

Open Access

INVITED REVIEW

Prostate Cancer

New developments in the treatment of castration resistant prostate cancer

Roxanne Wadia, Daniel P Petrylak

In the past 5 years, the treatment and understanding of metastatic castrate resistant prostate cancer (CRPC) have improved dramatically. Our understanding of the mechanisms of castration resistance has allowed for the development of new drugs to target prostate cancer, and our understanding of genetic mutations may give us new tools with which to more accurately diagnose and be able to predict the course of this heterogeneous disease. This article summarizes the recent advances in the understanding of the development of CRPC, as well as the new drugs and targets, which have evolved from this basic research.
Asian Journal of Andrology (2014) **16**, 555–560; doi: 10.4103/1008-682X.127824; published online: 08 April 2014

Keywords: castration resistant prostate cancer; abiraterone; enzalutamide; chemotherapy; docetaxel; cabazitaxel; PSMA antibody

DEVELOPMENT OF CASTRATE RESISTANT PROSTATE CANCER

When metastatic prostate cancer is first diagnosed, the initial line of therapy is androgen deprivation. The goal is to inhibit the ability of testosterone to bind to the androgen receptor (AR) which subsequently activates, dimerizes and acts as a ligand-dependent transcription factor for multiple genetic elements. The best characterized AR target gene is KLK2 which encodes prostate-specific antigen (PSA).[1] Androgen deprivation has been showed to trigger apoptotic regression in both benign and malignant prostatic epithelium.[2] On average patients develop castrate resistant prostate cancer (CRPC) after 18–24 months of androgen blockade, usually first exhibited by rising PSA despite serum testosterone <50. As PSA transcription is known to be regulated by AR, a rise in PSA is often the first harbinger of CRPC.

Although CRPC was previously felt to be hormonally independent, recent data implicate the continued activation of the androgen axis as a stimulus for growth. Xenograft studies have confirmed the central role of increased AR expression in CRPC development.[3] Development of CRPC occurs through two general mechanisms: alterations in the AR - allowing for deoxyribonucleic acid (DNA) transcription in the setting of low or no ligand and alterations in ligand production - creating a new source of androgens to bypass the blockade. Understanding the mechanisms of how CRPC develops is central to our development of new treatments.

AR itself functions as a ligand dependent transcription factor in the presence testosterone or dihydrotestosterone (DHT). It is present diffusely throughout the cytoplasm and is held in an inactive state. AR binds testosterone or its higher affinity ligand DHT, which testosterone is converted to in the prostatic epithelium or in the prostatic adenocarcinoma cells. Ligand binding to the AR results in the release of AR from its chaperone proteins and subsequent auto-dimerization. AR translocates to the nucleus where it binds DNA at specific sequences, known as "androgen responsive elements" (AREs) where it recruits co-activators that thus allow it to transcribe the AREs. AR antagonists are thought to result in a conformational change in the AR that leads to the binding of co-repressors instead of co-activators-resulting in repression of DNA transcription. Gonadotropin-releasing hormone (GNRH) agonists suppress the release of luteinizing hormone from the anterior pituitary preventing testosterone biosynthesis by Leydig cells within the testis and resulting to suppression of testicular testosterone synthesis, thus depriving the AR of its ligand, and preventing its activation.

There are multiple mechanisms to the development of resistance to AR antagonism and GNRH agonists.[1] Alteration of the AR itself; amplification, deregulation, mutation and post-translational modifications have all been characterized. The majority of CRPCs have been shown to have induction of AR messenger ribonucleic acid and protein expression with some CRPCs shown to have amplification and overexpression of the AR locus.[4–7] A transgenic model of mutant AR (AR-E231G) expression was found to have oncogenic transformation and subsequent development of metastatic disease substantiating that loss of AR regulation can in and of itself lead to the development of prostate cancer.[8] Mutations of the AR can lead to a change in the ligand binding domain leading to more promiscuous ligand binding, in some cases non-androgen steroid hormone. Treatment with AR antagonists like bicalutamide may provide selection pressure leading to the formation of these mutations in many cases.[9] Some alternative spliced ARs have deletion of the ligand binding domain, leading to an AR that is constitutively active, and upregulated in CRPCs.[10–12] Post-translational modifications to AR have been associated with development of CRPC, phosphorylated AR has been shown to be less responsive to androgen deprivation and *in vitro* studies have shown that tyrosine phosphorylated AR can act in a ligand-independent fashion.[13] Lastly, changes in AR co-activators and co-repressors are thought to play a significant role in AR disregulation. There is increased expression of a subset of AR co-activators in human CRPC cells, which may allow the AR receptor to transcribe genetic

Department of Medicine, Smilow Cancer Center, Yale University School of Medicine, Yale Comprehensive Cancer Center, New Haven, Connecticut, USA.
Correspondence: Dr. DP Petrylak (daniel.petrylak@yale.edu)
Received: 16 December 2013; Accepted: 12 February 2014

elements despite a low hormone environment.[14–16] Loss of co-repressors may also allow for aberrant signaling-converting AR antagonists into agonists.[17] It is likely that most CRPCs employ several of these mechanisms in simultaneously, as demonstrated by a 2001 study by Gregory *et al.*[18] in which recurrent tumors were shown to have increased AR expression as well as increased sensitivity to ligand.

Another mechanism through which CRPC develops is through the continued existence of testosterone and DHT despite androgen blockade. Multiple studies in the past 30 years have shown that despite castrate levels of testosterone in the serum of patients on systemic androgen suppression, the intra-prostatic androgen levels are adequate to activate the AR.[19] This evidence is supported by the activity of further androgen blockade through the second line anti-androgen therapy such as abiraterone acetate and enzalutamide to lead to PSA declines and regression of disease despite patients having "castrate level" serum androgens.

The presence of adequate levels of intra-prostatic DHT and testosterone in the prostate cancer cells demonstrates that there are alternative sources of testosterone. The adrenal gland is a well-known source of androgens, specifically dehydroepiandrosterone which is converted to testosterone in the prostate through a series of enzymatic reactions. In patients undergoing androgen deprivation therapy, the levels of adrenally derived androgens are still present in significant amounts.[20–22] Ketoconazole, is a weak inhibitor of CYP17 and 11β hydroxylase resulting in incomplete pharmacologic suppression of the adrenals use of ketoconazole in men with disease progression post-castration resulted 40%–50% response rates.[23] As the effect of ketoconazole does not completely suppress the adrenals, the degree of suppression was correlated with the response rate.[23]

Another source of intra-prostatic androgens post-castration is intracrine production in the prostate tissue itself. This occurs through two general mechanisms; *de-novo* synthesis from acetic acid and "back-door" synthesis which utilizes progesterone as a starting point and does not have testosterone as an intermediary in addition to the classical androgen biosynthesis pathway. Montgomery *et al.*[24] demonstrated that the steroidogenic enzymes required in the synthesis of testosterone and DHT from cholesterol precursors are all present within CRPC cells. Using a xenograft model, Locke *et al.*[2] incubated cells with ^{14}C labeled acetic acid, and demonstrated through high performance liquid chromatography (HPLC), the presence of labeled steroid precursors leading to detectable quantities of DHT. Furthermore, they were able to demonstrate that when ^{3}H progesterone was incubated with CRPC cells, there was conversion to DHT, with HPLC demonstrating the presence of intermediaries of both classic and the "back-door" synthesis pathways. Further evidence for CRPC reliance on intratumoral androgen production is the upregulation of androgen biosynthetic enzyme expression for both classical and backdoor pathways in metastatic CRPC tumors when compared to untreated primary prostate tumors. FASN, CYP17A1, HSD3B1, HSD17B3, CYP19A1, and UGT2B17 are all upregulated.[24] Relatively preserved progesterone levels in CRPC cells support its utilization by cancer cells to produce androgen.

DOCETAXEL

Docetaxel was the first cytotoxic chemotherapy approved for metastatic CRPC which showed a survival benefit. The previous standard of care was mitoxantrone combined with prednisone, reduced pain and improved quality of life scores without a survival advantage over prednisone alone. Docetaxel based therapy was compared to mitoxantrone in CRPC in two studies in 2004. In SWOG 99-16, docetaxel and estramustine were compared to mitoxantrone and prednisone in men with metastatic CRPC.[25] Patients were randomized to two treatment arms; estramustine days 1–5 and docetaxel day 2 of a 21 day cycle or mitoxantrone day 1 of a 21 day cycle. The primary endpoint of overall survival (OS) was reached with docetaxel extending median survival by approximately 2 months (17.5 *vs* 15.6 months). Secondary endpoints all showed significant improvement in the docetaxel arm including median time to progression (6.3 *vs* 3.2 months), and >50% PSA decline (50% *vs* 27%). The TAX327 study compared docetaxel given every 3 weeks or weekly docetaxel to mitoxantrone given every 3 weeks - all drugs were administered with prednisone.[26] A survival benefit was demonstrated only for every 3 week docetaxel over mitoxantrone. Median survival was significantly prolonged in the combined docetaxel group compared to the mitoxantrone group (18.9 *vs* 16.5 months). Among the secondary endpoints, reduction in pain reached statistical significance in the q3-week docetaxel but not in the weekly docetaxel group. PSA response rate was significantly higher in both docetaxel groups, although tumor response was equivalent. Quality of life scores were significantly improved in both docetaxel groups.

Approximately, one-quarter of the patients in the TAX327 study crossed over from one arm to another after disease progression. The median survival after crossover and PSA response was examined in these patients.[27] PSA response rate was higher for patients who received docetaxel post-mitoxantrone than patients who received mitoxantrone post-docetaxel (28% *vs* 15%). Median time to PSA progression was also improved in the group receiving docetaxel post-mitoxantrone (5.9 months *vs* 3.5 months). Despite these differences, there was no statistically significant difference in median survival post-crossover; median survival was 10 months and did not depend on the direction of the crossover. In addition, patients' response to the first line therapy did not predict their response to the second line therapy. Multiple trials have been performed with agents in combination with docetaxel unfortunately, none have shown improvement in OS compared with docetaxel and prednisone.[28–31]

SECOND GENERATION ANTI-ANDROGENS

Abiraterone

Abiraterone is an irreversible inhibitor of CYP17 that blocks androgen synthesis in the testis, adrenal glands and prostate. CYP17 or 17,20 lyase is responsible for conversion of 17α-hydroxyprogesterone to androstenedione, it plays an important role in both the classical and backdoor pathways of androgen biosynthesis. Use of abiraterone leads to undetectable levels of androgens in serum, and most significantly, undetectable intratumoral androgen levels-which was not seen with conventional androgen deprivation therapy.[32,33] Abiraterone has antitumor effect on both chemotherapy treated and chemotherapy-naïve patients with CRPC.

The first reported phase III study for abiraterone was in metastatic CRPC patients who had progressed post-docetaxel.[34] Patients were randomized 2:1 to abiraterone with 5 mg prednisone or placebo with 5 mg prednisone. The use of prednisone with abiraterone is necessary as abiraterone causes increases in cortisol levels leading to hypokalemia, hypertension and fluid retention. Patients with histologically or cytologically confirmed metastatic CRPC were eligible, these patients had prior treatment with docetaxel, had Eastern Cooperative Oncology Group (ECOG) performance status of two or less, and had serum testosterone levels of <50. Patients with neuroendocrine differentiation and those who had progression on ketoconazole therapy were not included. The primary endpoint evaluated was OS, secondary

endpoints included >50% PSA decline, time to PSA progression, and radiographic progression free survival (PFS). OS was significantly improved in patients who received abiraterone therapy (15.8 months *vs* 11.2 months) on placebo. OS was improved for the abiraterone group across all previously defined subgroups (age, ECOG status, pain scores, prior treatment, and disease extent) although not all of these differences were statistically significant. No significant difference was seen between the abiraterone group in the subgroup of patients who had progressed on docetaxel after receiving it for <3 months, these patients may represent a more aggressive subset of disease that is chemorefractory. Secondary endpoints also showed improvement in the abiraterone group; proportion of patients with >50% PSA decline (29.5% *vs* 5.5%), time to PSA progression (8.5 *vs* 6.6 months), median radiologic PFS (5.6 *vs* 3.6 months). The rates of significant adverse events were similar in both arms-fatigue being the most frequently reported.

Abiraterone was also investigated in a phase III trial in the chemotherapy-naïve patients.[35] Patients were randomized 1:1 to abiraterone plus 5 mg prednisone or placebo plus 5 mg of prednisone. The patients had histologically or cytologically confirmed adenocarcinoma, and PSA progression or radiographic progression in the setting of ongoing androgen deprivation therapy which was defined as serum testosterone <50 ng dl^{-1}, patients had ECOG performance status of 0 or 1. Patients on ketoconazole treatment for >7 days were excluded. Primary endpoints were radiographic PFS and OS. Secondary endpoints included time to opiate use for cancer-related pain, time to initiation of cytotoxic chemotherapy, time to decline in ECOG performance status and time to PSA progression. The radiographic PFS was significantly improved in the abiraterone group (16.5 *vs* 8.3 months). Median OS was not reached for the abiraterone group in the final analysis; the OS was 27.2 months for the placebo group and there was a 25% decrease in the risk of death in the abiraterone group, which showed a trend toward OS improvement. Secondary endpoints were all improved in the abiraterone arm; median time to opiate use for cancer related pain (not reached *vs* 23.7 months), median time to initiation of cytotoxic chemotherapy (25.2 *vs* 16.8 months), median time to decline in ECOG performance status (12.3 *vs* 10.9 months), and median time to PSA progression (11.1 *vs* 5.6 months) were all improved. Of note, at primary interim analysis the study was unblinded and crossover to the abiraterone arm was allowed-this could potentially explain the inability to meet the prespecified OS difference. The frequency of adverse events that necessitated stopping treatment was similar in both groups (19% abiraterone *vs* 12%) prednisone; the overall rate of adverse events was also similar with fatigue being the most commonly reports, liver function abnormalities and cardiac toxicities were more frequently reported for abiraterone.

The Ryan study was the first to show a second line hormonal manipulation for metastatic CPRC given prior to chemotherapy could improve radiographic PFS. At the time of progression, most patients were initiated on docetaxel treatment. Several *in vitro* studies have shown that, in addition to disrupting microtubule formation, that docetaxel may disrupt AR signaling.[36,37] Mezynski *et al*.[38] retrospectively analyzed whether treatment with abiraterone prior to the initiation of docetaxel affected the efficacy of docetaxel. A retrospective analysis of 54 patients enrolled in a phase I/II study of abiraterone was performed-35 of these patients had progression on abiraterone and were initiated on docetaxel. Docetaxel was started for progressive bony disease and worsening of pain. Eight of the 35 patients who progressed on abiraterone and transitioned to docetaxel were deemed "abiraterone-refractory" as they did not have a >50% PSA decline on abiraterone treatment. All the patients that were identified as abiraterone refractory, also failed to have a >50% PSA decline on docetaxel and were deemed to be "docetaxel refractory". Of the remaining 27 patients who had >50% PSA decline on abiraterone and were transitioned to docetaxel, nine had a >50% PSA decline on docetaxel. This study had borderline significance in rejecting the null hypothesis, indicating that there may be cross-resistance between abiraterone and docetaxel, which may be due to docetaxel's effect on AR signaling. There is also concern that stopping abiraterone may lead to the resurgence in intratumoral androgens, abrogating the potential antitumor activity of docetaxel. Patients who are both abiraterone and docetaxel refractory may represent a subset of disease that is androgen independent.

Transmembrane protease serine 2 (TMPRSS2)/ERG fusion

Chromosomal translocation resulting in the fusion product of transmembrane protease serine 2 (TMPRSS2) and members of the E-twenty-six (ETS) transcription factor family were found to be detected in approximately 50% of PSA-screened prostate cancer. They are very rarely detected in benign prostatic tissue.[39] In approximately 85% of cases, the TMPRSS2 was fused to the V-ETS erythroblastic virus ETS oncogene homolog ERG.[40–42] In xenograft models, the TMPRSS2:ERG translocation was associated with resurgent AR activity.[43] Analysis of prostatic intraepithelial neoplasia (PIN) versus invasive adenocarcinoma found that 20% of PIN lesions had ERG gene rearrangements compared to 50% of localized cancers, however all PIN lesions analyzed through fluorescence *in situ* hybridization (FISH) showed intermingling cancer foci expressing ERG rearrangements.[44,45]

Attard *et al*.[46] further characterized the fusion through studies utilizing FISH to identify the gene rearrangement. They examined the presence of the TMPRSS2-ERG gene rearrangement utilizing a break-apart FISH assay in a cohort of 445 prostate cancers from men who were managed conservatively. Three distinct FISH patterns were found; normal locus (N), rearrangement with preservation of the 5' and 3' sequences (Esplit), and loss of the 5' sequence with preservation of the 3' (Edel). While both Esplit and Edel corresponded to the TMPRSS2-ERG fusion, cancers exhibiting Edel were found to have a worse overall and cause-specific survival. Microarray analysis has shown that fusion positive and fusion negative prostate cancers have distinctive transcriptional profiles, insinuating that they represent biologically distinct disease.[47–49]

TMPRSS2:ERG fusion has potential utility as a biomarker of prostate cancer; both of presence of disease as well as potential aggressiveness. At present, the Gleason score is the only way that we can objectively characterize aggressiveness of disease and it is flawed in that it is a morphological test and interobserver variability is always present. ETS fusions can be detected through FISH at the chromosomal level, as well as by polymerase chain reaction (PCR)-based assays. Laxman *et al*.[50,51] showed detection of the TMPRSS: ERG fusion transcripts by reverse transcription-PCR in post-digital rectal examination (DRE) urine from men with known prostate cancer-it was detected in 42% of patients sampled. Four transcript biomarkers, collected from post-DRE urine had a higher positive predictive value than PSA or prostate cancer gene 3 for detection of prostate cancer in a sample of men with elevated PSAs undergoing biopsy or prostatectomy. Hessels *et al*.[52] demonstrated that while assessment for TMPRSS2-ERG fusion may have a high positive predictive value of 94%, its sensitivity is only 37%.

Enzalutamide

Enzalutamide, also known as MDV3100, is a pure AR antagonist and was initially found on a screen for non-steroidal anti-androgens against prostate cancer cells with AR overexpression. Unlike first generation anti-androgens such as bicalutamide and flutamide, it has no known

agonist activity.[53] Enzalutamide targets multiple steps in the AR signaling pathway. It not only blocks androgen binding to the AR but also prevents nuclear translocation and prevents AR from binding DNA and recruiting co-activators. It also binds to the AR with 5–8 fold higher affinity than bicalutamide.[53] Enzalutamide produced tumor shrinkage in xenograft models by suppressing tumor cell growth and inducing apoptosis, and inhibited the transcriptional activity of a bicalutamide resistant mutant AR.[53] Phase I/II trial looking at enzalutamide activity in patients with metastatic CRPC demonstrated clinical benefit.[54] Antitumor effect was noted in patients at all dose levels, 56% of all patients had >50% PSA decline, 22% had soft tissue response and 56% had stabilization of bony disease. Quantification of circulating tumor cells (CTC) was also assessed as an additional method of assessing medication effect. Initially 40% of patients had unfavorable CTC counts (>5 cells/7.5 ml of blood) these counts were reassessed after 12 weeks of therapy and 49% of these patients converted from unfavorable to favorable counts. The most common adverse event noted was fatigue, but two patients in the highest dose cohort had unwitnessed seizures.

The phase III trial compared enzalutamide to placebo in CRPC patients who were previously with docetaxel and had progressive disease by PSA measurement or radiographic progression.[55] Patients were randomized 2:1 to enzalutamide or placebo with the primary endpoint of OS. The study was stopped at the time of interim analysis when it was found that enzalutamide had a statistically significant OS compared to the placebo group (18.4 *vs* 13.6 months) with a 37% reduction in risk of death. Significant survival benefit was maintained across all subgroups (age, baseline pain intensity, geographic region, type of disease progression at entry). Secondary endpoints of >50% PSA reduction, soft tissue response rate, time to PSA progression, radiographic PFS, and time to first skeletal-related event were statistically superior in the enzalutamide arm. Enzalutamide had a lower incidence of grade 3 and 4 adverse events, but 5/800 enzalutamide patients had seizures compared to no seizures in the placebo group.

IMMUNOTHERAPEUTICS

Sipuleucel-T

Sipuleucel-T is the first immunotherapeutic that was approved for the treatment of CRPC. It is an autologous CD54 + dendritic cell vaccine directed again prostatic acid phosphatase (PAP), a marker expressed on prostate cancer cells. It is produced by harvesting dendritic cells from the patient and culturing them with recombinant granulocyte-macrophage colony-stimulating factor-PAP fusion protein. The activated cells are re-infused into the patient 4 days later and this process is repeated 3 times at 2 week intervals. The primed dendritic cells are then thought to activate T cells, which are targeted against the PAP antigen expressed on the prostate cancer cells. Sipuleucel-T was approved by the Food and Drug Administration (FDA) in 2010 for use in CRPC based on phase III data showing improved OS.[56] The IMPACT study was actually the third trial evaluating sipuleucel-T. It randomized patients 2:1 to receive sipuleucel-T or a placebo frozen dendritic cell product and was powered for OS benefit. Median survival was improved by 4.1 months with a 36.5 month median survival. At 3 years, the proportion of patients who had received sipuleucel-T and were alive was 38% higher than those who received placebo. Additional data showed that this benefit was present in almost every subset of patients; across Gleason score, PSA, extent of disease, age and laboratory values.[56]

Sipuleucel-T was approved based on the survival benefit seen in the IMPACT, without significant improvements in time to progression or PSA declines.[57,58] Two earlier trials failed to show a statistically significant difference in time to progression between the treatment and placebo arms, yet also demonstrated improved survival. This discordance between progression free and OS may be a common finding in immunotherapy trials for prostate cancer; a similar observation was noted when PROSTVAC, a PSA directed vaccine therapy, was compared to placebo in men with CRPC. Other clinical parameters do demonstrate a treatment effect in favor of sipuleucel-T. Sipuleucel-T is associated with longer time to disease-related pain, as well as a significant reduction in time to narcotic analgesic use, which may reflect the immune antitumor effect.[59] Additional studies have specifically examined markers of immune activation in patients receiving sipuleucel-T and found that markers of APC activation, antigen-specific T-cells, and peripheral immune response correlated with survival in patients receiving sipuleucel-T.[60] This enhanced population of antigen-specific T cells may be the mechanism of improved OS in sipuleucel-T.

Prostate specific membrane antigen (PSMA)

Another recent focus of immunotherapy in CRPC is prostate specific membrane antigen (PSMA). This transmembrane protein is expressed in 84%–100% of prostate cancers, restricted to the prostatic epithelium and upregulated after androgen suppression. Prostascint® is an FDA approved imaging agent that using ^{111}indium labeled antibody to an internal epitope of PSMA to image occult prostate cancer metastases. The development of multiple antibodies directed at external epitopes of PSMA, conjugated with radioactive isotopes or cytotoxic compounds, uses PSMA a therapeutic target. PSMA analog to digital converter with ADC is a fully human monoclonal antibody to PSMA that is conjugated to monomethyl auristatin E, an anti-tubulin. It was evaluated in the *in vitro* setting and found to be highly effective in killing cells with high PSMA expression.[61] Phase I studies in patient treated with prior chemotherapy have demonstrated clinical activity above 1.8 mg kg^{-1}. Phase II studies have recently been completed in docetaxel refractory[62] patients.

CONCLUSIONS

Significant gains in our understanding of the pathogenesis of prostate cancer and its treatment in the past 5 years. We have developed new agents that extend the life in CRPC. There remain many questions, particularly regarding the optimum timing and sequencing and combining of second generation anti-androgens and immunotherapeutics with conventional anti-androgen therapy and cytotoxic chemotherapy therapy. The overall therapeutic goal is to maximize treatment effect while minimizing toxicity.

COMPETING INTERESTS

Dr Daniel P Petrylak is the consultant of Bayer, Bellicum, Dendreon, Johnson and Johnson, Exelixis, Ferring, Millineum, Medivation and Pfizer. He has grant supports from Oncogenix, Progenies, Johnson and Johnson, Millineum, Celgene and Dendron.

REFERENCES

1 Knudsen KE, Penning TM. Partners in crime: deregulation of AR activity and androgen synthesis in prostate cancer. *Trends Endocrinol Metab* 2010; 21: 315–24.
2 Locke JA, Guns ES, Lubik AA, Adomat HH, Hendy SC, *et al*. Androgen levels increase by intratumoral de novo steroidogenesis during progression of castration-resistant prostate cancer. *Cancer Res* 2008; 68: 6407–15.
3 Chen CD, Welsbie DS, Tran C, Baek SH, Chen R, *et al*. Molecular determinants of resistance to antiandrogen therapy. *Nat Med* 2004; 10: 33–9.
4 Linja MJ, Savinainen KJ, Saramäki OR, Tammela TL, Vessella RL, *et al*. Amplification and overexpression of androgen receptor gene in hormone-refractory prostate cancer. *Cancer Res* 2001; 61: 3550–5.
5 Latil A, Bièche I, Vidaud D, Lidereau R, Berthon P, *et al*. Evaluation of androgen, estrogen (ER alpha and ER beta), and progesterone receptor expression in human prostate cancer by real-time quantitative reverse transcription-polymerase chain reaction assays. *Cancer Res* 2001; 61: 1919–26.

6 Ford OH 3rd, Gregory CW, Kim D, Smitherman AB, Mohler JL. Androgen receptor gene amplification and protein expression in recurrent prostate cancer. *J Urol* 2003; 170: 1817–21.
7 Edwards J, Krishna NS, Grigor KM, Bartlett JM. Androgen receptor gene amplification and protein expression in hormone refractory prostate cancer. *Br J Cancer* 2003; 89: 552–6.
8 Han G, Buchanan G, Ittmann M, Harris JM, Yu X, *et al*. Mutation of the androgen receptor causes oncogenic transformation of the prostate. *Proc Natl Acad Sci U S A* 2005; 102: 1151–6.
9 Steinkamp MP, O'Mahony OA, Brogley M, Rehman H, Lapensee EW, *et al*. Treatment-dependent androgen receptor mutations in prostate cancer exploit multiple mechanisms to evade therapy. *Cancer Res* 2009; 69: 4434–42.
10 Dehm SM, Schmidt LJ, Heemers HV, Vessella RL, Tindall DJ. Splicing of a novel androgen receptor exon generates a constitutively active androgen receptor that mediates prostate cancer therapy resistance. *Cancer Res* 2008; 68: 5469–77.
11 Hu R, Dunn TA, Wei S, Isharwal S, Veltri RW, *et al*. Ligand-independent androgen receptor variants derived from splicing of cryptic exons signify hormone-refractory prostate cancer. *Cancer Res* 2009; 69: 16–22.
12 Guo Z, Yang X, Sun F, Jiang R, Linn DE, *et al*. A novel androgen receptor splice variant is up-regulated during prostate cancer progression and promotes androgen depletion-resistant growth. *Cancer Res* 2009; 69: 2305–13.
13 Guo Z, Dai B, Jiang T, Xu K, Xie Y, *et al*. Regulation of androgen receptor activity by tyrosine phosphorylation. *Cancer Cell* 2006; 10: 309–19.
14 Xu J, Wu RC, O'Malley BW. Normal and cancer-related functions of the p160 steroid receptor co-activator (SRC) family. *Nat Rev Cancer* 2009; 9: 615–30.
15 Gregory CW, He B, Johnson RT, Ford OH, Mohler JL, *et al*. A mechanism for androgen receptor-mediated prostate cancer recurrence after androgen deprivation therapy. *Cancer Res* 2001; 61: 4315–9.
16 Agoulnik IU, Vaid A, Bingman WE 3rd, Erdeme H, Frolov A, *et al*. Role of SRC-1 in the promotion of prostate cancer cell growth and tumor progression. *Cancer Res* 2005; 65: 7959–67.
17 Burd CJ, Morey LM, Knudsen KE. Androgen receptor corepressors and prostate cancer. *Endocr Relat Cancer* 2006; 13: 979–94.
18 Gregory CW, Johnson RT Jr, Mohler JL, French FS, Wilson EM. Androgen receptor stabilization in recurrent prostate cancer is associated with hypersensitivity to low androgen. *Cancer Res* 2001; 61: 2892–8.
19 Geller J, Albert J, Nachtsheim D, Loza D, Lippman S. Steroid levels in cancer of the prostate–Markers of tumor differentiation and adequacy of anti-androgen therapy. *Prog Clin Biol Res* 1979; 33: 103–11.
20 Mizokami A, Koh E, Fujita H, Maeda Y, Egawa M, *et al*. The adrenal androgen androstenediol is present in prostate cancer tissue after androgen deprivation therapy and activates mutated androgen receptor. *Cancer Res* 2004; 64: 765–71.
21 Bélanger B, Bélanger A, Labrie F, Dupont A, Cusan L, *et al*. Comparison of residual C-19 steroids in plasma and prostatic tissue of human, rat and guinea pig after castration: unique importance of extratesticular androgens in men. *J Steroid Biochem* 1989; 32: 695–8.
22 Mohler JL, Gregory CW, Ford OH 3rd, Kim D, Weaver CM, *et al*. The androgen axis in recurrent prostate cancer. *Clin Cancer Res* 2004; 10: 440–8.
23 Robinson MR, Shearer RJ, Fergusson JD. Adrenal suppression in the treatment of carcinoma of the prostate. *Br J Urol* 1974; 46: 555–9.
24 Montgomery RB, Mostaghel EA, Vessella R, Hess DL, Kalhorn TF, *et al*. Maintenance of intratumoral androgens in metastatic prostate cancer: a mechanism for castration-resistant tumor growth. *Cancer Res* 2008; 68: 4447–54.
25 Petrylak DP, Tangen CM, Hussain MH, Lara PN Jr, Jones JA, *et al*. Docetaxel and estramustine compared with mitoxantrone and prednisone for advanced refractory prostate cancer. *N Engl J Med* 2004; 351: 1513–20.
26 Tannock IF, de Wit R, Berry WR, Horti J, Pluzanska A, *et al*. Docetaxel plus prednisone or mitoxantrone plus prednisone for advanced prostate cancer. *N Engl J Med* 2004; 351: 1502–12.
27 Berthold DR, Pond GR, de Wit R, Eisenberger M, Tannock IF, *et al*. Survival and PSA response of patients in the TAx327 study who crossed over to receive docetaxel after mitoxantrone or vice versa. *Ann Oncol* 2008; 19: 1749–53.
28 Tannock IF, Fizazi K, Ivanov S, Karlsson CT, Fléchon A, *et al*. Aflibercept versus placebo in combination with docetaxel and prednisone for treatment of men with metastatic castration-resistant prostate cancer (VENICE): a phase 3, double-blind randomised trial. *Lancet Oncol* 2013; 14: 760–8.
29 Fizazi KS, Higano CS, Nelson JB, Gleave M, Miller K, *et al*. Phase III, randomized, placebo-controlled study of docetaxel in combination with zibotentan in patients with metastatic castration-resistant prostate cancer. *J Clin Oncol* 2013; 31: 1740–7.
30 Kelly WK, Halabi S, Carducci M, George D, Mahoney JF, *et al*. Randomized, double-blind, placebo-controlled phase III trial comparing docetaxel and prednisone with or without bevacizumab in men with metastatic castration-resistant prostate cancer: CALGB 90401. *J Clin Oncol* 2012; 30: 1534–40.
31 Araujo JC, Mathew P, Armstrong AJ, Braud EL, Posadas E, *et al*. Dasatinib combined with docetaxel for castration-resistant prostate cancer: results from a phase 1-2 study. *Cancer* 2012; 118: 63–71.
32 Barrie SE, Potter GA, Goddard PM, Haynes BP, Dowsett M, *et al*. Pharmacology of novel steroidal inhibitors of cytochrome P450 (17) alpha (17 alpha-hydroxylase/C17-20 lyase). *J Steroid Biochem Mol Biol* 1994; 50: 267–73.
33 O'Donnell A, Judson I, Dowsett M, Raynaud F, Dearnaley D, *et al*. Hormonal impact of the 17alpha-hydroxylase/C (17,20)-lyase inhibitor abiraterone acetate (CB7630) in patients with prostate cancer. *Br J Cancer* 2004; 90: 2317–25.
34 Fizazi K, Scher HI, Molina A, Logothetis CJ, Chi KN, *et al*. Abiraterone acetate for treatment of metastatic castration-resistant prostate cancer: final overall survival analysis of the COU-AA-301 randomised, double-blind, placebo-controlled phase 3 study. *Lancet Oncol* 2012; 13: 983–92.
35 Ryan CJ, Smith MR, de Bono JS, Molina A, Logothetis CJ, *et al*. Abiraterone in metastatic prostate cancer without previous chemotherapy. *N Engl J Med* 2013; 368: 138–48.
36 Gan L, Chen S, Wang Y, Watahiki A, Bohrer L, *et al*. Inhibition of the androgen receptor as a novel mechanism of taxol chemotherapy in prostate cancer. *Cancer Res* 2009; 69: 8386–94.
37 Kuroda K, Liu H, Kim S, Guo M, Navarro V, *et al*. Docetaxel down-regulates the expression of androgen receptor and prostate-specific antigen but not prostate-specific membrane antigen in prostate cancer cell lines: implications for PSA surrogacy. *Prostate* 2009; 69: 1579–85.
38 Mezynski J, Pezaro C, Bianchini D, Zivi A, Sandhu S, *et al*. Antitumour activity of docetaxel following treatment with the CYP17A1 inhibitor abiraterone: clinical evidence for cross-resistance? *Ann Oncol* 2012; 23: 2943–7.
39 Tomlins SA, Rhodes DR, Perner S, Dhanasekaran SM, Mehra R, *et al*. Recurrent fusion of TMPRSS2 and ETS transcription factor genes in prostate cancer. *Science* 2005; 310: 644–8.
40 Mehra R, Tomlins SA, Shen R, Nadeem O, Wang L, *et al*. Comprehensive assessment of TMPRSS2 and ETS family gene aberrations in clinically localized prostate cancer. *Mod Pathol* 2007; 20: 538–44.
41 Han B, Mehra R, Dhanasekaran SM, Yu J, Menon A, *et al*. A fluorescence *in situ* hybridization screen for E26 transformation-specific aberrations: identification of DDX5-ETV4 fusion protein in prostate cancer. *Cancer Res* 2008; 68: 7629–37.
42 Kumar-Sinha C, Tomlins SA, Chinnaiyan AM. Recurrent gene fusions in prostate cancer. *Nat Rev Cancer* 2008; 8: 497–511.
43 Cai C, Wang H, Xu Y, Chen S, Balk SP. Reactivation of androgen receptor-regulated TMPRSS2: ERG gene expression in castration-resistant prostate cancer. *Cancer Res* 2009; 69: 6027–32.
44 Perner S, Mosquera JM, Demichelis F, Hofer MD, Paris PL, *et al*. TMPRSS2-ERG fusion prostate cancer: an early molecular event associated with invasion. *Am J Surg Pathol* 2007; 31: 882–8.
45 Mosquera JM, Perner S, Genega EM, Sanda M, Hofer MD, *et al*. Characterization of TMPRSS2-ERG fusion high-grade prostatic intraepithelial neoplasia and potential clinical implications. *Clin Cancer Res* 2008; 14: 3380–5.
46 Attard G, Clark J, Ambroisine L, Fisher G, Kovacs G, *et al*. Duplication of the fusion of TMPRSS2 to ERG sequences identifies fatal human prostate cancer. *Oncogene* 2008; 27: 253–63.
47 Tomlins SA, Mehra R, Rhodes DR, Cao X, Wang L, *et al*. Integrative molecular concept modeling of prostate cancer progression. *Nat Genet* 2007; 39: 41–51.
48 Tomlins SA, Laxman B, Varambally S, Cao X, Yu J, *et al*. Role of the TMPRSS2-ERG gene fusion in prostate cancer. *Neoplasia* 2008; 10: 177–88.
49 Iljin K, Wolf M, Edgren H, Gupta S, Kilpinen S, Skotheim RI, *et al*. TMPRSS2 fusions with oncogenic ETS factors in prostate cancer involve unbalanced genomic rearrangements and are associated with HDAC1 and epigenetic reprogramming. *Cancer Res* 2006; 66: 10242–6.
50 Laxman B, Tomlins SA, Mehra R, Morris DS, Wang L, *et al*. Noninvasive detection of TMPRSS2: ERG fusion transcripts in the urine of men with prostate cancer. *Neoplasia* 2006; 8: 885–8.
51 Laxman B, Morris DS, Yu J, Siddiqui J, Cao J, *et al*. A first-generation multiplex biomarker analysis of urine for the early detection of prostate cancer. *Cancer Res* 2008; 68: 645–9.
52 Hessels D, Smit FP, Verhaegh GW, Witjes JA, Cornel EB, *et al*. Detection of TMPRSS2-ERG fusion transcripts and prostate cancer antigen 3 in urinary sediments may improve diagnosis of prostate cancer. *Clin Cancer Res* 2007; 13: 5103–8.
53 Tran C, Ouk S, Clegg NJ, Chen Y, Watson PA, *et al*. Development of a second-generation antiandrogen for treatment of advanced prostate cancer. *Science* 2009; 324: 787–90.
54 Scher HI, Beer TM, Higano CS, Anand A, Taplin ME, *et al*. Antitumour activity of MDV3100 in castration-resistant prostate cancer: a phase 1-2 study. *Lancet* 2010; 375: 1437–46.
55 Scher HI, Fizazi K, Saad F, Taplin ME, Sternberg CN, *et al*. Increased survival with enzalutamide in prostate cancer after chemotherapy. *N Engl J Med* 2012; 367: 1187–97.
56 Kantoff PW, Higano CS, Shore ND, Berger ER, Small EJ, *et al*. Sipuleucel-T immunotherapy for castration-resistant prostate cancer. *N Engl J Med* 2010; 363: 411–22.
57 Higano CS, Schellhammer PF, Small EJ, Burch PA, Nemunaitis J, *et al*. Integrated data from 2 randomized, double-blind, placebo-controlled, phase 3 trials of active cellular immunotherapy with sipuleucel-T in advanced prostate cancer. *Cancer* 2009; 115: 3670–9.
58 Small EJ, Schellhammer PF, Higano CS, Redfern CH, Nemunaitis JJ, *et al*.

Placebo-controlled phase III trial of immunologic therapy with sipuleucel-T (APC8015) in patients with metastatic, asymptomatic hormone refractory prostate cancer. *J Clin Oncol* 2006; 24: 3089–94.

59 Small E, Higano C, Kantoff P, Whitmore J, Frohlich M, *et a*l. Time to disease-related pain after sipuleucel-T in asymptomatic patients with metastatic castrate resistant prostate cancer (mCRPC): results from 3 randomized phase III trials. In: European J Cancer. The Boulevard, Langford Lane, Kidlington, Oxford O×5 1GB, Oxon, England: Elsevier Sci. Ltd.; 2011. p. S484.

60 Sheikh NA, Petrylak D, Kantoff PW, Dela Rosa C, Stewart FP, *et al*. Sipuleucel-T immune parameters correlate with survival: an analysis of the randomized phase 3 clinical trials in men with castration-resistant prostate cancer. *Cancer Immunol Immunother* 2013; 62: 137–47.

61 Wang X, Ma D, Olson WC, Heston WD. *In vitro* and *in vivo* responses of advanced prostate tumors to PSMA ADC, an auristatin-conjugated antibody to prostate-specific membrane antigen. *Mol Cancer Ther* 2011; 10: 1728–39.

62 Petrylak DP, Kantoff PW, Mega AE, Vogelzang NJ, Stephenson S, *et al*. Prostate-specific membrane antigen antibody drug conjugate (PSMA ADC): a phase I trial in metastatic castration-resistant prostate cancer (mCRPC) previously treated with a taxane. *J Clin Onco*l 2013; 31 Suppl: [Abstr 5018].

How to cite this article: Wadia R, Petrylak DP. New developments in the treatment of castration resistant prostate cancer. *Asian J Androl* 08 April 2014. DOI:10.4103/1008-682X.127824. [Epub ahead of print]