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# Novel cytotoxic and biological agents for prostate cancer: Where will the money be in 2005?

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**Abstract**

In 2004, docetaxel-based chemotherapy became the first treatment capable of extending life in androgen-independent prostate cancer. The era of therapeutic nihilism in this disease has thus been put to rest and a broad range of agents is being tested with the goal of improving on the successes of 2004. Lessons learned from other tumour types will need to be applied to prostate cancer in order to harness the bounty of available ideas. Target amplification or activating mutations and not merely the presence of a target are likely to be important to the success of targeted agents. Thus, the promise of the current crop of targeted agents is most likely to be realised when pursued in the context of well-credentialed targets and tested in highly translational clinical trials that are capable not only of assessing tumour response, but also of evaluating the status of the targeted pathway. The most promising agents in clinical development are reviewed.


*Keywords:* Prostate cancer; Chemotherapy; Targeted therapy; Novel agents

## 1. Introduction

In 2004, docetaxel became the first drug to prolong survival of patients with androgen-independent prostate cancer (AIPC) and the long-held view that AIPC is an untreatable disease was put to rest. The advance seen with docetaxel is critically important because it demonstrates that progress in this disease is possible and it establishes a clear standard of care upon which future studies will be built. A substantial number of investigational agents, belonging to a wide range of drug classes and targeting a broad range of cancer pathways are in clinical development. Thus, the future has never been brighter in treating this disease, which is the second-leading cancer killer of men in the United States of America (USA) and in 2000 was the cause of more than 200,000 deaths worldwide.

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## 2. Current cytotoxic agents

The range of currently available cytotoxic agents has been summarised in detail by Bhandari, Petrylak and Hussain elsewhere in this volume. Active agents include mitoxantrone, the taxanes (paclitaxel and docetaxel, which seem to illustrate some schedule dependency) and estramustine [1–24]. The level of reported anti-cancer efficacy has depended somewhat on the case selection characteristics, the nature of 'response' (subjective or objective) and the use of surrogate markers of response.

## 3. New cytotoxic agents

### 3.1. SB-715992

Mitotic kinesins, which play essential roles in the assembly and function of the mitotic spindle, are

expressed preferentially in neoplastic cells. They represent novel targets for cancer treatment [25]. Inhibition of the mitotic kinesin spindle protein (KSP) results in formation of monopolar spindles within single cells and the induction of apoptosis in pre-clinical studies. SB-715992 is the first KSP inhibitor to enter clinical trials and has a broad spectrum of activity in pre-clinical models of cancer, including models that are refractory to chemotherapy and prostate cancer models. This agent has completed phase I evaluation and is entering phase II trials. The Southwest Oncology Group (SWOG) will be evaluating this agent in patients who have progressed on docetaxel-containing therapy.

### 3.2. Epothilones

The epothilones are a new class of cytotoxic agents with anti-neoplastic activity in pre-clinical models of a range of tumours insensitive or resistant to paclitaxel; their mechanism of action is microtubular stabilisation resulting in mitotic arrest [26]. EPO906 is a novel epothilone that is not a substrate for multidrug-resistance protein. A phase IIa trial of weekly EPO906 in 37 patients with previously-treated AIPC found that it was well tolerated; the most common adverse events were gastrointestinal. Twenty-two percent of patients responded by prostate-specific antigen (PSA) criteria and 4 out of 20 patients with measurable disease responded [27]. Two recent phase II studies demonstrate that the epothilone analogue BMS-247550 has activity in chemotherapy-naïve patients with metastatic AIPC. SWOG 0111 showed 41% PSA and 30% measurable disease response rates in patients with chemotherapy-naïve AIPC [28]. Another multi-institutional phase II study reported 56% PSA and 23% measurable disease response rates for BMS-247550 alone and 69% PSA and 44% measurable disease response rates for the combination of BMS-247550 with estramustine [29]. The most frequent grade 3 toxicities in both of these studies were fatigue and sensory neuropathy. These studies demonstrate activity for this agent, although its place in the treatment of AIPC is unclear in light of current docetaxel results. With the establishment of docetaxel-based chemotherapy as front-line therapy, suggestive evidence that there is some degree of non-cross-resistance with epothilones becomes increasingly relevant [30].

### 3.3. Satraplatin

Satraplatin is a novel oral platinum complex that has shown activity against AIPC in cisplatin-resistant human tumour lines and in phase I trials. Satraplatin plus prednisone was compared with prednisone alone in 50 chemotherapy-naïve AIPC patients. The satraplatin arm had a better progression-free survival (median 5.2 months *versus* 2.5 months, $P = 0.023$) and a higher frequency of PSA response (33.3% *versus* 8.7%; $P = 0.046$) [31]. A phase III trial of satraplatin plus prednisone versus prednisone as second-line chemotherapy is underway.

### 3.4. Amonafide

Amonafide is a synthetic imide anti-neoplastic agent with DNA intercalative properties that demonstrated significant activity in pre-clinical studies and some activity in phase I trials, including at least one partial response in a patient with prostate cancer [32]. The drug is extensively metabolised and detected in plasma and urine. Its toxicity has previously been correlated to the formation of an active metabolite, *N*-acetyl-amonafide. In phase I studies using various administration schedules of amonafide, myelosuppression was the dose limiting toxicity. A phase II trial of amonafide at a dose of 225 mg/m$^2$ i.v. daily for 5 d was conducted by SWOG. Forty-three evaluable patients with measurable AIPC were treated. The most common toxicities were haematological including leucopaenia (72%), granulocytopaenia (32.6%) and thrombocytopaenia (44.2%). There were no complete responses and 5 partial responses, giving an overall response rate of 12% [33].

Subsequent work determined that *N*-acetyltransferase enzyme polymorphisms play an important role in the metabolism of amonafide (fast acetylators are at great risk of haematological toxicity from amonafide therapy). Ratain and colleagues [34] proposed a pharmacodynamic model based on acetylator phenotype using caffeine as a probe to optimise amonafide dosing. Observations from a phase II trial of amonafide in patients with metastatic breast cancer that response rates appeared higher in patients experiencing more severe myelosuppression led to the hypothesis that optimisation of amonafide dosing might yield evidence of higher anti-tumour activity [35]. Using a modification of Ratain's pharmacodynamic model, a phase I/II trial of NAT2 phenotype-based dosing of amonafide is ongoing in patients with metastatic AIPC treated with up to one prior systemic chemotherapy regimen.

## 4. Nuclear receptor ligands

### 4.1. Peroxisome proliferators-activated receptor gamma (PPARγ) ligands

Peroxisome proliferators-activated receptor gamma (PPARγ) ligands are members of the nuclear receptor superfamily of ligand-dependent transcription factors and play a central role in lipid metabolism and adipocyte regulation. PPARs are widely expressed by prostate cancer cells. Synthetic PPARγ agonists inhibit prostate cancer growth *in vitro* [36]. The thiazolidinediones (including troglitazone, rosiglitazone and piaglitazone)

are a class of synthetic ligands for PPARγ that have been developed primarily for the treatment of insulin-resistant type II diabetes mellitus. A phase II study of troglitazone in patients with asymptomatic advanced prostate cancer included 12 men with androgen-dependent disease and 29 men with AIPC. In the androgen-dependent group, 1 of 12 patients had a >50% decrease in PSA and 3 of 12 patients had a <50% decrease. In the AIPC group, no patient had a >50% decrease in PSA but 4 of 29 patients had a <50% decrease [37]. A second randomised, placebo-controlled trial of rosiglitazone was initiated. A total of 105 men with rising PSA after local therapy were randomised with a primary end-point of post-treatment change in PSA doubling time (PSADT). There was no difference between the 2 arms in the primary efficacy analysis. Complicating analysis was the higher-than-expected (40%) response rate in the placebo arm [38,39]. Given these results, it is unlikely that PPARγ ligands alone will play a role in prostate cancer therapy. However, as the role of this receptor in prostate cancer progression is better understood, rationally designed combination of drugs that include PPARγ ligands may emerge.

## 5. Calcitriol

Calcitriol, the most active metabolite of vitamin D, has significant anti-neoplastic activity in a broad range of pre-clinical cancer models including *in vitro* and *in vivo* activity in several models of prostate cancer [40–46]. Several mechanisms of activity have been proposed. These include inhibition of proliferation associated with cell cycle arrest and, in some models, differentiation, reduction in invasiveness and angiogenesis, and induction of apoptosis. Further synergistic and/or additive effects with cytotoxic chemotherapy, radiation and other cancer drugs have been reported. A contemporary comprehensive review of spectrum and mechanisms of activity is available [47].

Significantly supraphysiological concentrations of calcitriol are required for anti-neoplastic effects. The development of high-dose regimens, made possible by intermittent dosing of calcitriol [48,49], has led to several clinical trials in prostate cancer. Single agent weekly calcitriol in hormone-naïve prostate cancer appeared to lengthen PSADT in an uncontrolled study [50]. Weekly calcitriol yielded encouraging phase II results when added to docetaxel. Eighty-one percent of 37 patients had confirmed >50% reduction in serum PSA in a single institution trial [51]. These results were encouraging relative to historical experience with single agent docetaxel and led to the development of a randomised trial of docetaxel with calcitriol or placebo. This trial (dubbed ASCENT) is using DN-101, a highly concentrated formulation of calcitriol developed specifically for oncology. ASCENT accrual has been completed and results should provide robust information about the potential of calcitriol to enhance chemotherapy for prostate cancer. Studies of calcitriol with dexamethasone, paclitaxel and carboplatin are underway as well [52].

The development of calcitriol analogues that might have anti-neoplastic activity but cause less hypercalcaemia has been proposed as another strategy to target the vitamin D receptor for cancer treatment. Several hundred such analogues have been synthesised [53–55]. Several analogues, dosed daily, have entered clinical trials. Two of 25 AIPC patients had objective partial responses to 1-alpha-hydroxyvitamin $D_2$ [56]. Further studies of calcitriol analogues in prostate cancer are underway.

## 6. Growth factor receptor antagonists

### 6.1. Epidermal growth factor receptor

The epidermal growth factor (EGF) system has been implicated as one of the mitogenic signals that may drive progression of AIPC. EGF receptor (EGFR) signalling pathways may be involved in prostate cancer invasion and angiogenesis [57,58]. Cetuximab is an anti-EGFR monoclonal antibody with high affinity that prevents activation of receptor tyrosine kinase. In pre-clinical studies cetuximab has demonstrated activity in a PC-3M-LN4 orthotopic model of AIPC, both as a single agent and in combination with paclitaxel [59]. A phase Ib/IIa study of cetuximab and doxorubicin in 18 patients with AIPC documented stabilisation in PSA for a median of 4 months in 5 patients (28%) [60]. Gefitinib (Iressa) is an oral small molecule EGFR tyrosine kinase inhibitor that has demonstrated clinical utility in non-small cell lung cancer [59,61]. However, a phase II trial of gefitinib in AIPC has preliminarily found infrequent PSA responses and early progression in many patients [62]. Thus the EGF receptor is a target that has yet to be vetted in a clinical trial and its role in the future of prostate cancer therapy remains to be determined.

### 6.2. HER-2

Trastuzumab is a humanised monoclonal antibody that targets the extracellular domain of HER-2 and confers a survival benefit when combined with chemotherapy in patients with metastatic breast carcinoma that overexpresses HER-2 [63]. Subsequent analyses have suggested that most of the activity of this agent is seen in patients with HER-2 gene amplification and fluorescent *in situ* hybridisation has largely replaced immunohistochemistry as a tool for identifying breast cancer patients who are likely to benefit from trastuzumab. HER-2 expression in prostate cancer, as determined by

immunohistochemistry, has produced very heterogeneous results [64,65]. In one series of 70 patients with metastatic disease, the overall rate of prostatic-tissue sections presenting positive HER-2 immunostaining was 64.3% [66] and in another study all 39 patients with prostate cancer showed positive staining by immunohistochemistry [67]. Conversely, two other series identified HER-2 positivity in 40% (32 of 79) and 0% (0 of 40) of patients with prostate cancer [68,69]. A phase II trial in patients with progressive prostate cancer found that trastuzumab alone produced no responses in 23 patients with either HER-2 negative (17 patients) or HER-2 positive (6 patients) disease as determined by immunohistochemistry. Additionally, four patients with HER-2 positive disease received combination therapy with trastuzumab and paclitaxel; two patients had a partial response by PSA and two patients had disease progression [65]. A second phase II trial of trastuzumab and docetaxel in HRPC was closed early when only 7/72 patients were found to have HER-2 overexpression by immunohistochemistry and none of the 4 patients treated with trastuzumab alone demonstrated response [64]. Thus it appears that trastuzumab is unlikely to gain a foothold in prostate cancer therapy.

### 6.3. PDGF receptor

Platelet-derived growth factor (PDGF) and its receptor (PDGF-R) have been implicated as both paracrine and autocrine mediators of prostate cancer progression [70,71]. The PDGF-R is expressed in 80% of AIPC lesions [72]. Imatinib (Gleevec) is a specific inhibitor of the BCR-ABL, c-kit and PDGF receptor tyrosine kinases. In a mouse model of prostate cancer bone metastases, PDGF-R inhibition by imatinib resulted in preservation of bone structure, significant tumour growth inhibition, a significant increase in apoptosis of malignant cells and a decrease in lymph node metastases [61]. Therefore, PDGF-R would appear to be a promising target in prostate cancer therapy. A small study of 15 patients with AIPC and painful bony metastases treated with imatinib and zoledronic acid failed to demonstrate PSA response and had no palliative or clinical activity [73]. A phase II trial of imatinib as a single agent in 17 patients with hormone-naïve PSA progression after local therapy demonstrated biochemical stable disease in 6 patients (35%) [74]. At the present time, it is not clear whether this class of agents will prove useful. Although little single agent activity was seen, imatinib may prove more useful in combination therapy. A study at MD Anderson is examining imatinib in combination with docetaxel. As with all targeted therapies, it will be important that future studies include determination of PDGF-R expression whenever feasible, so that clinical outcomes can be interpreted in the context of knowledge about the status of the target.

### 6.4. IGF receptor

Interest in the IGF system and prostate cancer was stimulated by epidemiological investigations that suggest a relationship between elevated IGF-1 levels and prostate cancer risk [75,76] and recognition that signalling through the IGF-1 receptor inhibits apoptosis. Targeting the IGF signalling system is therefore a promising strategy in prostate cancer prevention and therapy. Agents that target the IGF system are in pre-clinical development.

## 7. Anti-angiogenic agents

### 7.1. Thalidomide

As is the case for most solid tumours, the recruitment of blood vessels is an important step in progression and metastasis of prostate cancer. Inhibition of this complex process is an attractive approach to treatment and many anti-angiogenic agents are currently in clinical development. Thalidomide is a sedative, anti-inflammatory and immunosuppressive agent that blocks the activity of angiogenic agents including bFGF, VEGF and IL-6. The limb defects attributed to its teratogenic effects have been postulated to be secondary to an inhibition of blood vessel growth in the developing foetal limb bud. A recent clinical trial of thalidomide for AIPC randomised 63 patients to a low-dose arm of 200 mg daily or a high-dose arm, escalating to the highest tolerated dose (up to 1200 mg) [77]. Only 13 patients were treated on the high dose arm that was terminated early due to a combination of lack of efficacy and poor tolerance. In all, 9 patients (15%) showed a PSA decline of over 50%, all of whom were in the low-dose arm. A second phase II study of low-dose thalidomide in 20 patients with AIPC identified 3 men (15%) with a decline in serum PSA of at least 50% [78]. A randomised phase II trial of docetaxel with or without thalidomide 200 mg daily in 53 patients with chemotherapy-naïve AIPC reported that 19 of 36 patients (53%) in the combination arm and 6 of 17 (35%) receiving docetaxel alone had a PSA decrease of at least 50%. Combination therapy also resulted in improved progression-free survival (5.9 *versus* 3.7 months) and overall survival at 18 months (68.2% *versus* 42.9%) [79]. Therapy was relatively well tolerated, although 12 of the first 43 patients in the combined treatment group developed thromboembolic events; all subsequent patients received prophylactic anticoagulation with low molecular weight heparin and no further thromboembolic events were noted. Thus thalidomide appears to have modest single-agent activity and its activity with docetaxel appears additive.

### 7.2. Lenalidomide

CC-5013 (lenalidomide, Revlimid) is an immunomodulatory thalidomide analogue, which in animal models is both anti-angiogenic and non-teratogenic [80].

A phase I trial in patients with melanoma and other solid tumours provided evidence that CC-5013 can boost Th1-type cellular immunity. Figg and colleagues are currently conducting a phase I trial of CC-5013 in AIPC patients [81].

### 7.3. Bevacizumab

Bevacizumab (Avastin) is a recombinant, humanised anti-vascular endothelial growth factor (VEGF) monoclonal antibody that blocks the binding of VEGF to its receptors. Recently approved in the United States for first-line use in metastatic colorectal cancer, bevacizumab is also being examined in prostate cancer. CALGB 90006 is an ongoing phase II trial of bevacizumab, docetaxel and estramustine in patients with AIPC. Initial results include a confirmed >50% decline in PSA in 13 of 20 patients (65%) with sufficient data and a partial response in 9 of 17 patients (53%) with measurable disease [82].

### 7.4. Other

PTK787/ZK 222584 is an oral angiogenesis antagonist that inhibits all known VEGF receptors. A phase I study in 23 patients with AIPC showed that 3 (13%) had >40% reductions in PSA [83]. A recent randomised phase II trial of two doses of the matrix metalloproteinase inhibitor BMS-275291 in 80 patients with AIPC resulted in no responders [84].

## 8. Proapoptotic agents

### 8.1. Bcl-2 antisense

The mitochondrial-associated protein Bcl-2 confers resistance to apoptosis and is overexpressed in AIPC [85,86]. G3139 (oblimersen sodium) is an 18-mer phosphorothioate antisense oligonucleotide directed to the first six codons of the initiating sequence of the human bcl-2 mRNA [87]. In pre-clinical prostate cancer models, Bcl-2 antisense oligonucleotides have inhibited expression of Bcl-2, delayed development of androgen independence and enhanced the effects of chemotherapy [88–91]. A phase II study of G3139 and docetaxel in 31 men with metastatic AIPC (8 had received prior chemotherapy) yielded a confirmed >50% PSA reduction in 15 patients (48%) and a PR in 4 out of 15 patients (27%) with measurable disease. Toxicities included grade 3–4 neutropaenia in 42% of patients [92]. As this was a small study, it is difficult to determine whether this level of activity represents an improvement from that expected with docetaxel alone.

### 8.2. Selective apoptotic anti-neoplastic drugs (SAANDs)

Exisulind (sulindac sulphone), the oxidative metabolite of sulindac, is a member of a class of novel drugs that inhibit growth and induce apoptosis in prostate cancer cell lines by specifically inhibiting cyclic GMP phosphodiesterases but not cyclooxygenase-1 or -2 [93,94]. Goluboff and colleagues [95] randomised 96 patients with rising PSA levels after radical prostatectomy to 12 months of exisulind 250 mg orally or placebo twice daily and demonstrated that exisulind significantly suppressed the increase in PSA in all patients and prolonged the PSA doubling times (PSADT) in high-risk patients. Reduction in the PSADT persisted through 24-month follow-up on an open-label extension [96]. Additionally, pre-clinical studies have suggested synergistic interactions between exisulind and several chemotherapeutic agents [97–100] and a phase II study examining the combination of exisulind and docetaxel in men with AIPC has completed accrual [101].

CP-461 is an exisulind analogue with broad antitumour activity. In a phase I study, 21 patients with a range of solid tumours (not including prostate cancer) received CP-461 twice daily for 28 d. Therapy was well tolerated and 4 patients exhibited disease stability after two cycles of treatment [93]. A phase II study of CP-461 in prostate cancer is underway.

### 8.3. Clusterin antisense oligonucleotide

Clusterin is a survival gene whose expression increases markedly in response to androgen-deprivation therapy [102]. The expression of clusterin confers a chemotherapy-resistant phenotype, probably due to reduction in treatment-induced apoptosis [103,104]. Oligonucleotide antisense molecules to clusterin have been developed and have entered clinical trials in prostate cancer [105]. Development in combination with chemotherapy is expected.

## 9. Other novel targeted agents

### 9.1. Bortezomib

Bortezomib (PS-341, Velcade) is a novel boronic acid dipeptide that inhibits 26S proteasome activity. Although this agent has been most extensively studied in multiple myeloma, there is some intriguing evidence of its potential utility in prostate cancer. Several investigators have demonstrated that bortezomib is active in

pre-clinical models of human prostate cancer. Ikezoe and colleagues demonstrated that bortezomib blocks the androgen receptor signalling pathway and induces growth arrest and apoptosis in LNCaP cells [106]. Williams and colleagues found that bortezomib appeared to have anti-tumour activity in both LNCaP and PC-3 lines, but with distinctly different underlying mechanisms. The LNCaP lines demonstrated reduced microvessel density and VEGF secretion and high levels of apoptosis, while the PC-3 lines had direct increases in tumour cell death [107].

Papandreou and colleagues conducted a phase I trial of bortezomib in 48 patients with AIPC. Doses ranging from 0.13 to 2 mg/m$^2$ were administered weekly × 4, with cycles repeated every 5 weeks. Dose-limiting toxicity was noted in 2 of 5 patients treated at the 2 mg/m$^2$ dose level (grade 3 diarrhoea in both patients, grade 3 syncope and hypotension in 1 patient). The maximally tolerated dose (MTD) recommended with this schedule was 1.6 mg/m$^2$. Twenty-four patients were treated at dosages in the range of the MTD ($\geqslant$1.45 mg/m$^2$). Two patients (8%) demonstrated a $\geqslant$50% decline in PSA and 1 of 9 (11%) patients with measurable nodal disease achieved a partial response [108]. Based on preclinical data suggesting potential synergism with chemotherapy, phase II trials of bortezomib in combination with docetaxel were initiated [109–112]. Dreicer and colleagues conducted a phase I/II trial (the phase II portion consisted of three separate cohorts of patients treated at various dosages) of bortezomib plus docetaxel in 100 patients with metastatic AIPC. Data from the phase I portion of the study and the low expanded cohort were presented recently. Docetaxel was given at 25, 30, 35 and 40 mg/m$^2$ (cohorts 1–4) i.v. over 30 min on days 1 and 8. For the first 4 cohorts, bortezomib was given at a fixed dose of 1.3 mg/m$^2$ i.v. push on days 2 and 9, while a fifth cohort evaluated a higher dose of bortezomib at 1.6 mg/m$^2$ with 40 mg/m$^2$ of docetaxel, with treatment in all cohorts repeated every 21 d. In the phase I portion of the study no maximally tolerated dose was determined. A total of 32 patients were enrolled in the low expanded cohort. Twenty-one patients had prior chemotherapy, 11 with a taxane-containing regimen. Therapy was well tolerated; anaemia was the most common grade 3 event (2 if 32 patients). Six of 25 patients (24%) evaluable for response had a confirmed PSA decrease of $\geqslant$50% from baseline. Three of 13 (23%) patients with measurable disease achieved a partial response [113].

### 9.2. Atrasentan

Endothelin-1 is a potent vasoconstrictor produced by prostate cancer and appears to have a role in prostate cancer progression and morbidity. Atrasentan is an oral, highly selective endothelin receptor antagonist. In a phase III, placebo-controlled trial that involved 809 patients with metastatic AIPC, atrasentan did not prolong time to clinical progression (HR 1.14, 95% CI 0.98–1.34) in an intent-to-treat analysis. Atrasentan therapy was associated with smaller increases in serum PSA and markers of bone turnover. A modest delay in time to clinical progression (HR 1.26, 95% CI 1.06–1.50) was noted in the protocol-compliant patients [114].

An intent-to-treat meta-analysis that included this study as well as a prior randomised phase II study demonstrated a delay in disease progression (HR 1.19, $P = 0.013$), but the overall clinical benefit was small. These studies are characterised by rapid early progression of a subset of patients, which contributed to the difficulty detecting a benefit in the individual trials. A phase III study of this agent in earlier stages of AIPC will probably determine its fate.

## 10. Immunotherapeutic agents

### 10.1. GM-CSF

Granulocyte-macrophage colony stimulating factor (GM-CSF) is a cytokine that regulates the proliferation and differentiation of myeloid precursor cells and enhances the function of both mature granulocytes and mononuclear phagocytes. GM-CSF can influence the recruitment, activation and survival of macrophages and dendritic cells. Additionally, experimental evidence suggests that GM-CSF may increase the efficiency of tumour antigen presentation and the subsequent activation of tumour-specific cytotoxic T lymphocytes [115,116]. Small and colleagues administered GM-CSF (250 μg/d daily × 2 weeks followed by three times a week administration) to patients with metastatic AIPC to evaluate their hypothesis that apoptotic tumour cells could provide a source of tumour antigen, which might lead to T-cell cross-priming by dendritic cells. Therapy was well tolerated and a small subset of the patients experienced a greater than 50% decline in PSA, with one patient demonstrating both a prolonged PSA response and objective improvement in bone scan imaging [117]. Dreicer and colleagues treated 16 patients with advanced prostate cancer (7 hormonally-naïve and 9 androgen-independent) with GM-CSF administered subcutaneously at 250 μg thrice weekly for up to 6 months. Although no patient achieved an objective response, 6 patients demonstrated a 10–15% decline in their baseline PSA that was maintained during the entire treatment period. Five of these 6 patients demonstrated a rise in their PSA following study completion. Therapy was well tolerated, with only one grade 3 event, which was probably not treatment-related [118].

### 10.2. GVAX

GVAX cancer vaccines are comprised of tumour cells that have been genetically modified to secrete GM-CSF. GVAX was studied in a phase II dose escalation trial. Nineteen patients were treated at the highest dose. Of these patients, 6 (32%) had modest declines in serum PSA and 87% demonstrated immune response [119]. Phase III studies of GVAX are planned.

### 10.3. Provenge

Provenge consists of autologous dendritic cells loaded *ex vivo* with a recombinant fusion protein of prostatic acid phosphatase (PAP) linked to GM-CSF. In early studies, Provenge was shown to induce immune responses to PAP in 38% of patients and occasional PSA reductions in excess of 50% [120]. A single durable response has been reported [121]. In the initial randomised study in AIPC, Provenge failed to improve progression-free survival, but a benefit in progression-free survival and overall survival was seen in a subset of patients whose Gleason score was less than 8 [122]. As a result, Provenge is now being evaluated in patients with metastatic AIPC that at initial diagnosis did not exceed Gleason 7. It is also being tested in a placebo-controlled randomised study in patients with a rising serum PSA after prostatectomy.

### 10.4. Vaccinia virus/fowlpox virus

Various investigators have pursued immunotherapy with viral vectors since they can mimic natural infection and produce potent immune responses. Investigators from the Eastern Cooperative Oncology Group (ECOG) have recently reported a phase II trial designed to evaluate the tolerability and feasibility of a prime/boost vaccine strategy using vaccinia virus and fowlpox virus expressing human PSA in patients with PSA progression following definitive local therapy [123]. A total of 64 eligible patients with PSA progression (hormone-naïve) following definitive local therapy (radical prostatectomy/radiotherapy) were randomly assigned to receive four vaccinations with fowlpox-PSA (rF-PSA), three rF-PSA vaccines followed by one vaccinia-PSA (rV-PSA), or one rV-PSA vaccine followed by three rF-PSA vaccines with the primary endpoint being PSA response at 6 months. Therapy was well tolerated and 45% of men remained free of PSA progression at 19.1 months. There was a trend favouring the treatment group that received a priming dose of rV-PSA. Although no significant increases in anti-PSA antibody titres were detected, 46% of patients demonstrated an increase in PSA-reactive T cells [123]. ECOG is currently developing a randomised phase II trial of fowlpox-PSA with and without GM-CSF.

### 10.5. MVA-MUC1-IL2 vaccine

Over-expression, non-polarity and under-glycosylation of the mucin glycoprotein MUC1 is associated with many epithelial neoplasms, making MUC1 a potential target of vaccine immunotherapy. MUC1 expression has been reported in 60–90% of prostate cancer specimens evaluated [124,125]. Modified vaccinia ankara (MVA) is a highly attenuated vaccinia virus that is non-propagative in most mammalian cells. TG4010 is a recombinant MVA expressing MUC1 and IL-2 that demonstrated an excellent safety profile in phase I evaluations. Interim results from a randomised phase II trial of two different schedules of TG4010 in patients with hormone-naïve biochemical failure have been reported recently. Eligible patients were required to have a PSA doubling time of less than 10 months, with an absolute value of 2.0 ng/ml or greater without evidence of metastatic disease. Patients were randomly assigned to receive either weekly injections of $10^8$ plaque-forming units for the first 6 weeks then injections every 3 weeks until week 36 or progressive disease, or the same dosage administered every 3 weeks until week 36 or progressive disease. At the time of the interim assessment, 29 patients had been treated on study, with a significant change in PSA doubling time from pre-treatment assessment favouring arm 1 (weekly therapy) [126].

### 10.6. J591 monoclonal antibody

Prostate-specific membrane antibody (PSMA) is a cell-surface glycoprotein expressed (not secreted) in both benign and malignant prostate tissue. Unlike the imaging agent indium-111 capromab penditide (a radiolabelled murine antibody that binds to an intracellular epitope of PSMA), J591 binds the extracellular PSMA epitope and has been de-immunised by engineering it into a human immunoglobulin G1 [127]. Bander and colleagues reported a phase I trial of naked antibody and then initiated a series of early trials with various radioisotopes [128]. Investigators at Cornell performed a phase I trial of yttium-90-labelled J591 in 29 patients with advanced AIPC [127]. Dose limiting toxicity was seen at 20 $mCi/m^2$ with two patients experiencing thrombocytopaenia with non-life-threatening bleeding episodes requiring platelet transfusions. Four patients were retreated without evidence of a human–antihuman antibody response. Anti-tumour activity was seen, with two patients having greater than 50% decline in PSA lasting 8 and 8.6 months and this was associated with measurable disease responses in soft tissue metastases. Two other phase I studies are in progress, one using the beta-emitting radiometals lutetium-177 and the second using the chemotherapy agent maytansinoid-1 linked to J591 [128].

## 11. Conclusion

The first decade of the 21st century is an unprecedented time in prostate cancer research. The therapeutic nihilism of previous decades has given way to considerable enthusiasm. A growing understanding of the molecular events that lead to prostate carcinogenesis has led to the identification of a large number of potential therapeutic targets. Lessons learned from other tumour types will need to be applied to prostate cancer to best harness the bounty of available ideas. Experience from breast cancer, lung cancer, gastrointestinal stromal tumour and chronic myelogenous leukaemia suggests that the mere presence of a target is unlikely to be sufficient. The identification of target amplification or activating mutations has produced the most promising new treatment strategies in other tumour types and is likely to be equally important in prostate cancer. Thus, the current armamentarium of novel agents offers much promise, and this promise is most likely to be realised when pursued in the context of well-credentialed targets and tested in highly translational clinical trials that are capable of not only assessing tumour response, but also evaluating the status of the targeted pathway.

## Conflict of interest statement

Dr. T. Beer has declared a fiscal conflict of interest with respect to reporting of data on the use of calcitriol for prostate cancer.

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