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Clinical Oncology Update: Prostate Cancer

Androgens and Prostate Cancer: Biology, Pathology and Hormonal Therapy

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INTRODUCTION—PATHWAYS OF ANDROGEN PRODUCTION AND MECHANISMS OF ACTION

ANDROGENS ARE required for the development and maintenance of both the normal prostate and prostate cancer [1]. Ninety to ninety-five per cent of circulating androgens are produced in the testis. Luteinising hormone-releasing hormone (LHRH) is produced in the hypothalamus and secreted into the hypophyseal portal circulation to reach the anterior pituitary (Figure 1). There, LH (luteinising hormone) is released which stimulates the Leydig cells of the testis to produce testosterone. Testosterone is taken up from the circulation by the prostate gland epithelium, where it is converted to the most active androgen, 5α-dihydrotestosterone, by 5α-reductase. The testes also contribute approximately 30% of plasma androstenedione, although the adrenals are the main source of this hormone [2].

In a second pathway, corticotrophin-releasing hormone from the hypothalamus stimulates the release of andrenocorticotrophic hormone (ACTH) from the pituitary. This in turn causes the release of adrenal androgens: androstenedione, dehydroepiandrosterone (DHEA) and its sulphate. The adrenal precursor, DHEA is converted into testosterone and then 5α-dihydrotestosterone by the pathway shown, either in the plasma or in the prostate itself, as the prostate is able to take up and metabolise adrenal androgens directly [3]. After castration (medical or surgical), plasma testosterone levels fall to one tenth of normal, but the amount of 5α-dihydrotestosterone detectable in prostatic tissue may be 40–50% of normal [4]. This fact underlies the rationale for total androgen blockade (see below).

In addition to the above pathways, growth hormone (GH) and prolactin influence the production of androgens from the testis and adrenal gland. Prolactin acts with LH to stimulate testosterone secretion from the testes. It also increases the release of adrenal androgens and has a direct mitogenic effect on prostatic cells [5]. GH is present in human prostatic tumours and prostatic tissue contains specific binding sites for GH [6]. Its full role in normal prostate and prostatic cancer is not known.

Tissue growth factors, such as epidermal growth factor (EGF), insulin-like growth factor (IGF), transforming growth factors (TGFα, β) and fibroblast growth factor (FGF) also affect growth of the normal prostate, and alterations in their secretion or their receptors may be important in prostatic cancer [7, 8]. They are often secreted from epithelial and stromal cells in a paracrine or autocrine manner, regulating both these cellular compartments. Carruba and associates [9] have recently demonstrated that growth of an androgen independent prostatic cell line, PC3, was stimulated by TGFα and inhibited by TGFβ, a commonly inhibitory factor. In contrast, hormone responsive LNCaP was not sensitive to either growth factor. IGF-1 stimulates the growth of prostatic cells and its actions may be modified by IGF binding proteins which are secreted by prostate epithelial cells [10]. Neuroendocrine cells are also present in the normal prostate, in the ductal and acinar epithelium. They contain a variety of secretory granules including serotonin, somatostatin, bombesin and calcitonin, secreted by endocrine, paracrine, neurocrine or lumencrine mechanisms. They play an important role in regulation of growth, differentiation and secretory function of the prostate gland. In prostatic carcinoma, focal neuroendocrine differentiation is common, correlates with higher grade, and neuroendocrine cells may be hormone resistant [11]. The hormonal control of growth in both normal prostate, and in prostate cancer, is therefore complex, with androgens having a predominant, but not exclusive role.

5α-dihydrotestosterone, once formed in the prostate epithelial cell, binds to the androgen receptor in the nucleus (Figure 2). This receptor also binds testosterone, with 4- to 5-fold less affinity. The androgen receptor belongs to the steroid receptor superfamily of ligand-dependent transcription factors, including receptors for oestrogen, progesterone, glucocorticoids, thyroid hormone and retinoic acid. Target cells for androgens are located in many parts of the body, with the highest concentration of androgen receptors in the male accessory organs, for example male hair follicles, genital skin, epididymis, seminal vesicles, prostate, as well as in

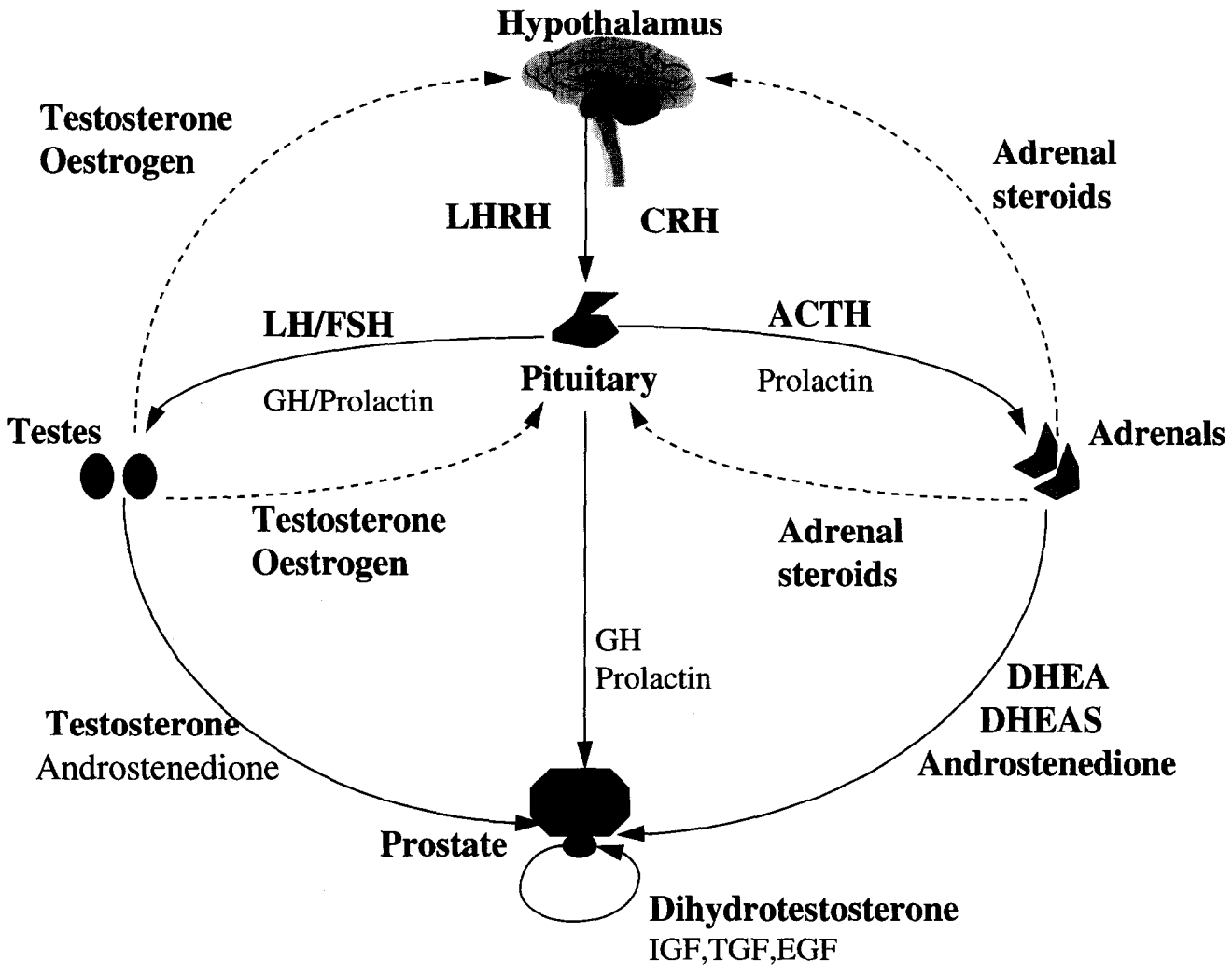

Figure 1. Hormonal pathways involving the prostate gland. Broken lines indicate negative feedback. LHRH, luteinising hormone-releasing hormone; ACTH, andrenocorticotrophic hormone; LH, luteinising hormone; FSH, follicle-stimulating hormone; GH, growth hormone; CRH, corticotrophin-releasing hormone; DHEA/DHEAs, dehydroepiandrosterone.

the hypothalamus. Other tissues such as skeletal muscle, liver and heart, have lower concentrations of receptors [2].

The androgen receptor consists of 919 amino acids, and is structurally organised in three domains: an N-terminal transactivating domain (exon A), a DNA-binding domain (exons B and C), containing two zinc fingers which bind to androgen responsive elements—specific sequences of DNA—and a C-terminal hormone binding domain (exons D–H) (Figure 3). The gene is located at Xq11-12, and is inherited in an X-linked manner [12].

In the absence of hormone, the androgen receptor is found mainly in the nucleus, associated with heat-shock proteins. Once 5α-dihydrotestosterone binds to the hormone binding domain, the heat-shock proteins dissociate, the receptor is hyperphosphorylated and dimerises. The DNA binding domain then binds to the androgen-responsive elements of genes such as that encoding the prostate-specific antigen (PSA), or other androgen responsive genes involved in the control of prostate cell division, interacting with other transcription factors to regulate their expression.

EFFECTS OF ANDROGEN DEPRIVATION ON NORMAL PROSTATE—APOPTOSIS

Apoptosis, or programmed cell death, is an energy dependent process of biochemical and morphological changes resulting in DNA and cellular fragmentation, producing cell death without an inflammatory cell influx. It may be activated either by sufficient injury to the cell by agents such as radiation, cytotoxic drugs, viruses, etc. or by changes in the pattern of endogenous signals such as hormones and growth factors.

Often, proliferating cells undergo apoptotic death if their progression through the cell cycle is perturbed, for example by the presence of DNA damage, triggering a rise in p53 protein, and arrest in G1. However, Furuya and associates [13] have demonstrated that prostatic glandular cells, after androgen ablation, undergo apoptosis in G0 without recruitment into G1, and that an increase in p53 is not involved in this process. Other evidence suggests that the cytoreductive effect of androgen ablation is attributable to the withdrawal of an antagonistic action of androgen on apoptosis [14]. Thus, androgen-sensitive cells require the presence of androgen to continue their normal function, and if androgen

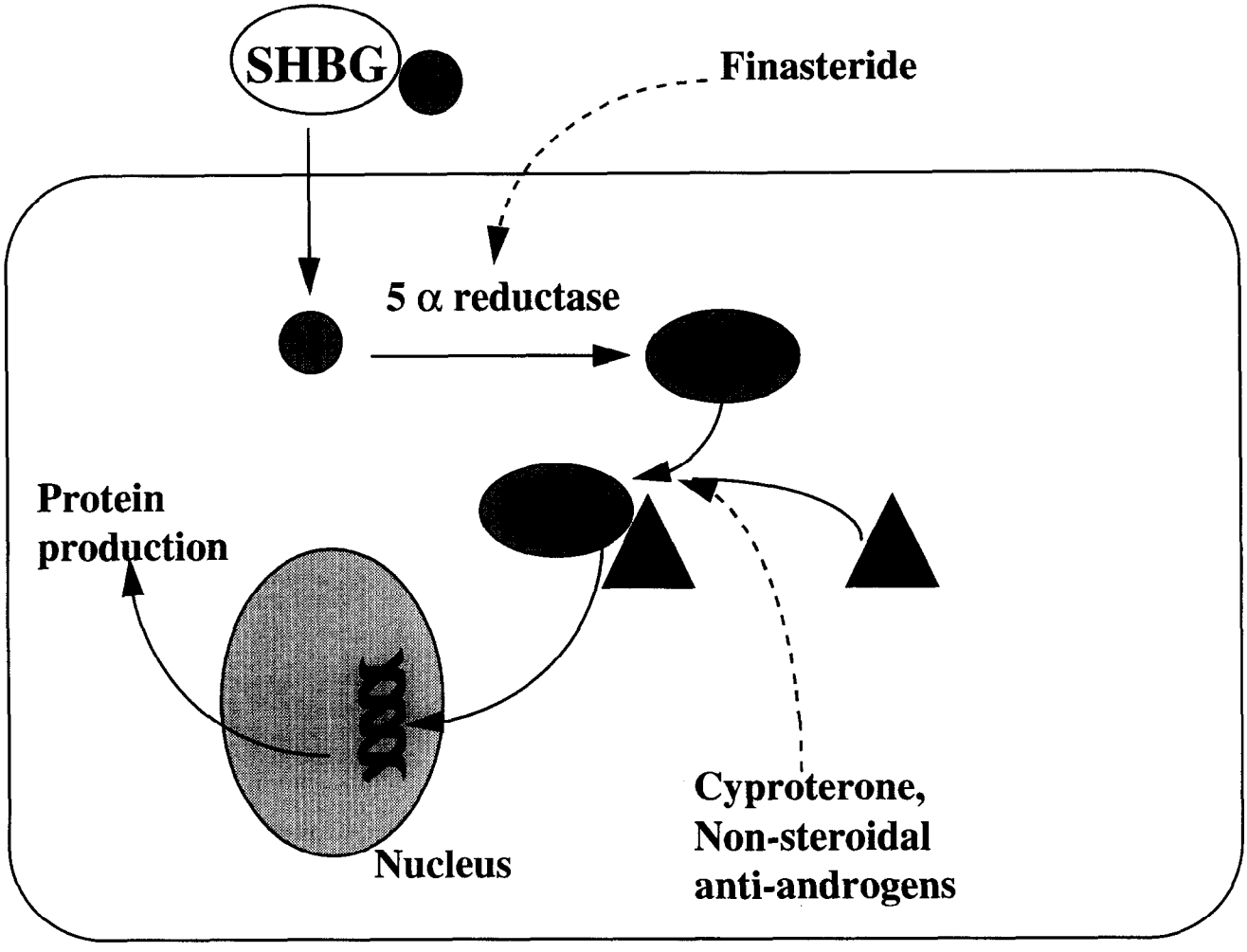

Figure 2. Intracellular androgen action. Broken lines indicate inhibitory effect. T, testosterone; DHT, dihydrotestosterone; R, androgen receptor; SHBG, sex hormone binding globulin.

is withdrawn they may undergo apoptosis, even if the cells are not in the proliferative cell cycle.

Bcl-2 protein is known to inhibit apoptosis, and there is evidence from the prostate cancer cell line PC-3, that altered levels of bcl-2 may be related to a differential susceptibility to apoptosis, explaining why the normal reaction to androgen deprivation is lost in some prostate cancer cells [15].

MECHANISMS OF ACTION OF HORMONAL MANIPULATION

There are various ways of producing regression of prostatic cancer by means of hormonal manipulation. Table 1 summarises these, with advantages and side-effects of each method.

Gonadal androgen ablation

Removing or reducing androgens of gonadal origin may be achieved by surgical castration, administration of oestrogens or inhibition of the synthesis and release of LH and FSH (follicle-stimulating hormone) by gonadotrophin releasing hormone agonists. Androgen deprivation has been the mainstay of treatment of metastatic disease since the 1940s [16]. Approximately 80% of patients with metastatic disease have a symptomatic and objective response,

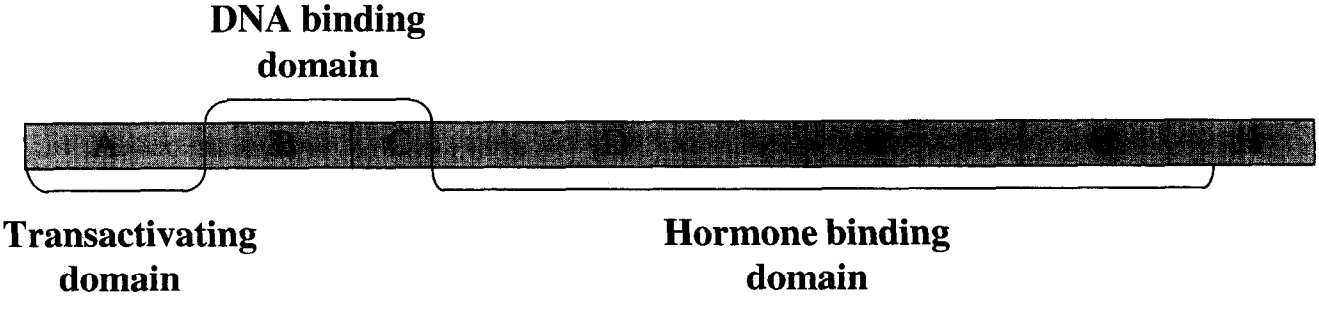

Figure 3. The androgen receptor gene.

Table 1. Summary of hormonal treatments for prostate cancer

	Advantages	Disadvantages
Gonadal androgen ablation		
Surgical castration	-rapid fall of androgens -patient compliance	-psychological effects -impotence, hot flushes
Diethylstilboestrol	-cheap	-gynaecomastia, impotence -risk of myocardial infarction, stroke, thrombo-embolism
LHRH agonists	-reversibility -depot injections aid compliance	-initial tumour flare -impotence, hot flushes
Anti-androgens		
Steroidal		
Cyproterone	-cheap	-small increased risk of thrombosis -impotence -hepatic toxicity
Megestrol acetate		-less effective than cyproterone
Non-steroidal	*-all preserve potency*	
Flutamide		-diarrhoea -gynaecomastia, hot flushes
Nilutamide	-once daily dose	-hot flushes -light-dark adaptation disorders -alcohol intolerance
Bicalutamide	-once daily dose	-mild gynaecomastia
Steroidal synthesis inhibitors	*-useful as second line*	
Ketoconazole		-impotence, gynaecomastia -severe gastrointestinal symptoms, hepatotoxicity -adrenal insufficiency
Aminoglutethimide		-anorexia, nausea, vomiting -depression -transient rashes, oedema -adrenal insufficiency
Hydrocortisone	-cheap -simple	-adrenal insufficiency if stopped suddenly
5α-reductase inhibitors		
Finasteride		-reduced potency -less effective than medical/surgical castration
New agents	*-may be effective in 'hormone refractory' disease*	
Suramin		-fatigue -renal impairment -axonal neuropathy
Somatostatin analogues		-gastro-intestinal toxicity

including reduction in size of primary and metastatic tumours, producing relief of bone pain and reduction in urinary obstructive symptoms, as well as a fall in serum PSA, and improvement in general well-being. The median duration of response is 15–18 months, with median survival from the time of treatment of symptomatic metastases of approximately 2.5 years. The rates and duration of response are similar for castration, oestrogens and LHRH agonists [17, 18].

Bilateral orchidectomy was the original form of androgen ablation and remains a standard procedure [16]. It has the advantage of rapid reduction in circulating testosterone by 95% within 3 h of surgery, and subsequent rapid symptomatic improvement [19]. It also has a low cost, and there is no problem with patient compliance. Disadvantages include the psychological effects of losing the testes, which may be lessened by the subcapsular technique, surgical morbidity, which is usually small, and the irreversibility of hormone ablation. In addition, the physical side-effects include impotence, hot flushes and mild breast tenderness. There is a permanent rise in LH and FSH levels due to loss of negative feedback on the pituitary.

Oestrogens, such as diethylstilbestrol (DES), act by a central mechanism, blocking gonadotrophin releasing hormone secretion through negative feedback. This causes a fall in LH and therefore a reduction in serum testosterone levels to castration levels in 10–14 days [20]. There may also be a direct effect on Leydig cells: oestrogen can competitively bind to the androgen receptor, although with less than 1% of the affinity of 5α-dihydrotestosterone [21]. If treatment is continued for 3 years or more, testosterone levels remain suppressed for many months after stopping oestrogens, despite slowly increasing LH levels. Oestrogens also stimulate the production of sex hormone binding globulin, which binds circulating androgens thus reducing the amount of free androgen available to bind the androgen

receptor. DES, at a dose of 3 mg/day, effectively suppresses testosterone to castration levels. Disadvantages of oestrogens include feminisation, with gynaecomastia, impotence and altered fatty tissue deposition in virtually 100% of patients. Gynaecomastia may be prevented by prophylactically irradiating the nipples. Of more concern are the cardiovascular side-effects. Oestrogens cause metabolic changes, including a rise in serum triglycerides and high-density lipoproteins, as well as alterations in coagulation factors, with increases of factors VII, VIII and fibrinogen. This leads to an increased risk of myocardial infarction (3%), stroke (2%), deep venous thrombosis (4–5%) and pulmonary embolism (4%) [22]. A daily dose of 1 mg of DES is as clinically effective as 3 mg, has fewer cardiovascular side-effects, but does not reliably reduce serum testosterone to castration levels. The addition of warfarin 1 mg daily reduces the risk of thrombotic events.

Use of LHRH agonists such as leuprolide, buserelin or goserelin, at a sustained and constant level, rather than the normal pulsatile manner of release of LHRH from the hypothalamus, initially stimulates LH and testosterone release to 1.40–1.70 times basal levels within several days. However, there is then downregulation of the LHRH receptors in the pituitary, producing castration levels of testosterone by 1 month. The initial increase in testosterone may cause a tumour 'flare', with increase in pain from bony metastases, or worsening urinary symptoms in 5–10% of patients [23]. This is of particular concern when there is imminent cord compression or nodal obstruction of a ureter: in these situations, initial treatment with LHRH agonists alone is contra-indicated. Tumour flare can be prevented with the addition of direct anti-androgenic drugs such as cyproterone or flutamide in the initial few weeks of treatment. Other side-effects include impotence, hot flushes and loss of libido, which are reversible on stopping treatment. These agents have the advantage of convenience, with 1 monthly and now 3 monthly depot injections available.

Anti-androgens

Anti-androgenic drugs compete with androgens for binding to the androgen receptor, and when bound, have an antagonistic action. They are classified as steroidal or non-steroidal compounds. Cyproterone acetate and megestrol acetate are synthetic steroidal anti-androgenic drugs that also have progesterone activity, including inhibition of pituitary gonadotrophin secretion, thus reducing plasma testosterone levels. In addition, they block 5α-reductase. Cyproterone has fewer cardiovascular side-effects than DES, but there is still a small increase in risk of thrombotic events [24]. Side-effects include depression, dyspnoea, impotence and loss of libido. Recently, the Committee of Safety on Medicines (U.K.) has reported 96 cases (33 fatal) of hepatic reactions to cyproterone, out of about 720 000 prescriptions. The reactions included hepatitis, cholestatic jaundice and hepatic failure, and arose mainly in those treated for several months [25]. It is, therefore, recommended that cyproterone should only be used for short courses to cover the initial tumour flare with LHRH agonists, and that liver function tests should be checked before and during treatment. Megestrol acetate has been shown to be less efficacious than cyproterone or DES, and is not widely used [24].

Non-steroidal anti-androgenic drugs—flutamide, bicalutamide (casodex) and nilutamide—block cellular binding of androgens to the androgen receptor in peripheral tissues, and act centrally, opposing the negative feedback of testosterone on the pituitary. Testosterone levels may therefore rise, so theoretically this may reduce their efficacy, but there is no evidence of this from clinical trials. Indeed, the maintenance or rise in testosterone levels means fewer side-effects on sexual function, with approximately 50–70% of patients maintaining potency and libido. In previously untreated patients, flutamide alone had a 87% response rate, with 85% of men who were potent prior to treatment retaining potency [26]. Gynaecomastia occurs, but less frequently than with DES therapy, as the rise in LH also produces a rise in oestrodiol levels, and there is also a rise in prolactin levels with these agents.

Flutamide is a derivative of toluidine, and is reduced to hydroxyflutamide, the active metabolite after absorption. The usual dose is 250 mg 8 hourly. Its most common side-effect is diarrhoea, but it may also cause nausea, gynaecomastia and rarely, altered liver function [26]. Nilutamide is structurally similar to flutamide. It has a longer half-life and, therefore, the advantage of once daily dosing (300 mg/day). Its side-effects include hot flushes, nausea, light–dark adaptation disorders, alcohol intolerance and rarely interstitial pneumonitis [27]. Bicalutamide is a new, potent, long-acting non-steroidal anti-androgen, with a half-life of 5–7 days. Libido and sexual function are usually maintained and hot flushes are infrequent. It does not cause gastrointestinal disturbance, hepatic impairment or visual symptoms. However, mild gynaecomastia or breast tenderness occurs in about half of patients [28]. The comparison of this agent with flutamide may have been affected by its much shorter duration of clinical application and experience.

Inhibition of adrenal steroid synthesis

Agents which directly inhibit androgen synthesis include aminoglutethimide and ketoconazole. Ketoconazole is an antifungal agent and imidazole derivative, which interferes with gonadal and adrenal androgen synthesis as well as the synthesis of cholesterol, inhibiting cytochrome P450 enzymes via interaction at the haem iron site [29]. Testosterone is suppressed to a greater extent than cortisol as there is selective inhibition of C17-20 lyase at low doses, thus blocking the conversion of 17-OH-pregnenolone to DHEA, and 17-OH-progesterone to androstendione (see Figure 4) [30]. The hormonal changes produced by ketoconazole are dose-dependent and fully reversible, but there are wide variations in serum levels of testosterone and ketoconazole in patients receiving the same dose [31]. This may relate to its short half-life: recovery from suppression begins 8 h after an oral dose and is complete by 24 h. With a dose of 400 mg 8 hourly, castration levels of testosterone are initially achieved. However, there is a rise in LH due to lack of negative feedback, and this leads to a progressive rise in testosterone levels [32]. Despite this, good clinical responses occur. Side-effects include impotence, gynaecomastia, severe gastrointestinal disturbances, hepatotoxicity (10% have transient abnormalities in liver function, <1% have more severe hepatic injury), and signs of adrenal insufficiency requiring hydrocortisone replacement. Compliance with the high-dose regimen and 8 hourly doses is also a

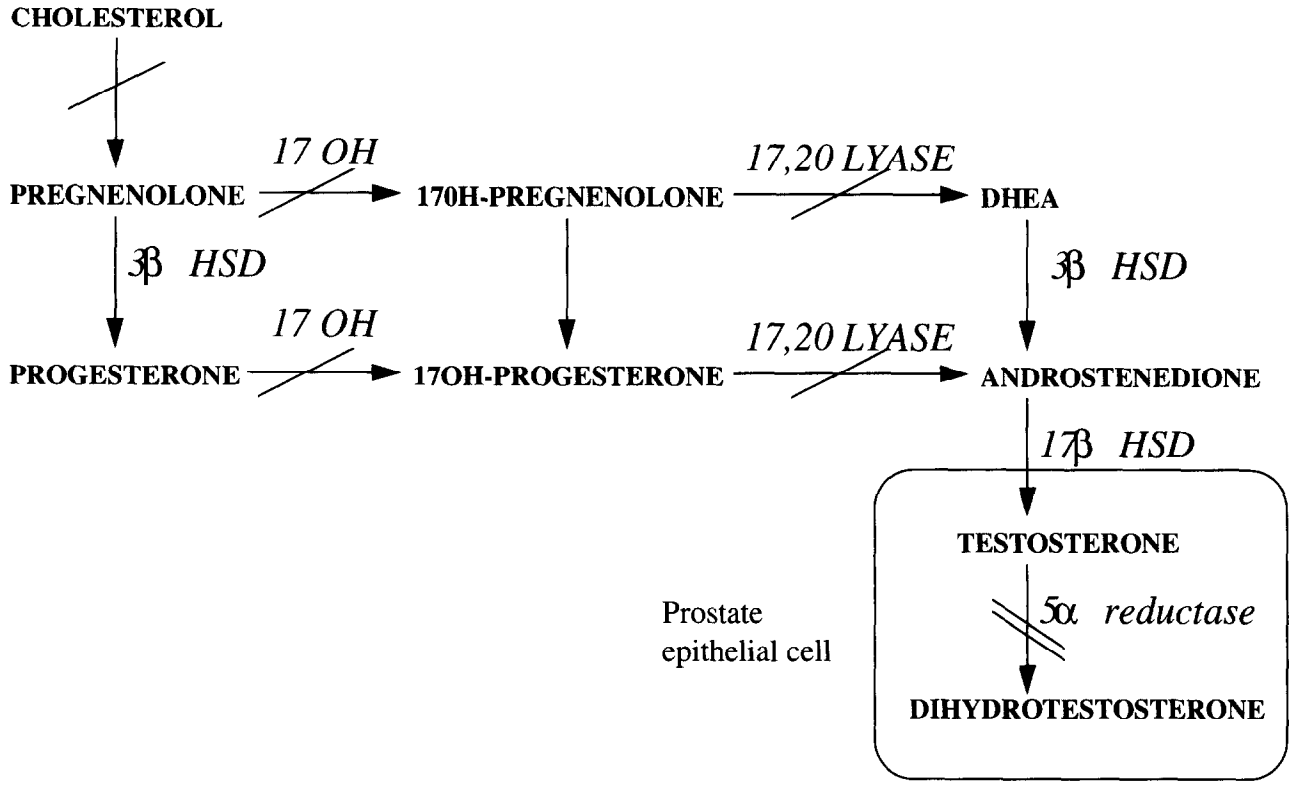

Figure 4. Pathways of androgen production. Single line represents site of ketoconazole inhibition, double line site of finasteride action. HSD, hydroxysteroid dehydrogenase; OH, hydroxylase; DHEA, dehydroepiandrosterone.

problem, leading to large diurnal variations in serum testosterone in most patients.

Aminoglutethimide blocks the conversion of cholesterol to pregnenolone by inhibiting 20-24 desmolase (see Figure 4). This reduces synthesis of glucocorticoids, mineralocorticoids and sex steroids by the adrenal glands, which may necessitate replacement with hydrocortisone. Endocrine studies have cast doubt on whether the clinical effect of aminoglutethemide is due to androgen suppression [33]. It is also a potent inhibitor of aromatase, the enzyme mediating the conversion of androgens to oestrogens, and of prostaglandin synthetase, so its mode of action remains unclear. Side-effects include nausea and vomiting, anorexia, depression, peripheral oedema and transient skin rashes [34].

Selective aromatase inhibitors, such as 4-OH-androstendione, have been used in the palliation of patients with advanced hormone resistant prostatic cancer [35]. In a group of heavily pretreated patients, 72% had a subjective response, with a similar number experiencing a transient tumour flare. Oestrodiol levels fell with treatment, but the mechanism for the beneficial effect remains unclear.

After orchidectomy or treatment with LHRH agonists, the addition of hydrocortisone acts by suppression of ACTH secretion through negative feedback on the pituitary [36]. The production of adrenal androgens is, therefore, reduced. This simple manoeuvre has fewer side-effects than ketoconazole or aminoglutethimide, and is much cheaper than the non-steroidal anti-androgens.

Inhibition of 5α-reductase

Finasteride is a competitive 5α-reductase inhibitor that does not bind to the androgen receptor. It reduces prostatic concentrations of dihydrotestosterone more than castration, but there is a 7-fold rise in prostatic testosterone, although total prostatic androgen is decreased [37]. When used in men with metastatic prostate cancer, Presti and associates [38] demonstrated a significant reduction in PSA, but the size of the reduction did not approach that of medical or surgical castration.

New agents

Suramin is an investigational agent that binds to heparin-binding growth factors, and has been shown to reduce levels of IGF-I and -II. It also has adrenolytic activity. Myers and associates reported its use in hormone refractory prostate cancer, with one third of patients having objective responses, some lasting more than a year [39]. A further phase I trial had a 75% response rate with dose-limiting toxicity of fatigue, renal impairment and axonal neuropathy [40]. Given the effects of growth hormone on prostate cell growth and the presence of neuroendocrine cells in normal prostate as well as in prostatic carcinoma, it is not surprising that somatostatin and its analogues inhibit the growth of prostate cancer cells. This has been demonstrated in rats bearing the androgen independent Dunning prostate tumours [41, 42]. Phase I–II studies in patients with hormone refractory prostate cancer have had conflicting results, with some reporting reduction or stabilisation of PSA in 30–40% [43], but others showing no response and significant gastro-intestinal toxicity [44, 45].

MECHANISMS OF HORMONE INDEPENDENCE

If other causes of death do not intervene, all metastatic prostatic cancers eventually become hormone independent,

with progression of disease despite continued androgen ablation. Possible mechanisms by which hormone independence occurs are discussed below. There is likely to be heterogeneity of these mechanisms both within individual tumours and between different patients.

Variation in androgen receptor content in prostate tumours

As the androgen receptor mediates the action of androgens, it was thought that its content in tissue might correlate with response to hormonal therapy. However, assessment using binding assays, or immunohistochemical staining has not demonstrated a lack of androgen receptor in hormone refractory disease, nor a prediction about individual tumour behaviour [46].

Mutations of androgen receptor gene

One mechanism for androgen independence may be mediated by mutations in the androgen receptor gene which allow the receptors to continue to stimulate the growth of prostate cancer cells in the absence of any hormone binding, or when alternative steroid hormones bind.

A mutation which does alter steroid binding and response to anti-androgens has been found in the human prostate cancer cell line LNCaP [47]. This produces a receptor with high binding affinities for progesterone, cyproterone, oestradiol, flutamide and nilutamide. When bound to the receptor, they produce the same effects as androgens. If similar mutants exist *in vivo*, it would explain the phenomenon of the flutamide withdrawal response [48].

In primary prostate cancer, mutations in the androgen receptor gene were thought to have been unusual [49–52]. More frequent mutations have been found when large parts of the cDNA of the gene have been sequenced. Tilley and associates [53] found mutations in approximately half the prostate cancers examined. Taplin and colleagues [54] have demonstrated somatic point mutations in the androgen receptor gene in 5 of 10 patients with metastatic androgen-independent prostate cancer; all in the hormone binding domain. These mutations were not detectable in specimens of the primary cancers. Two of the mutant receptors could be activated by progesterone and oestrogen.

Alteration in expression of the androgen receptor

Reduced androgen receptor messenger RNA has been found in prostate cancer cell lines [55]. However, levels of expression of the androgen receptor detected by semiquantitative PCR were high in those patients with androgen-independent disease examined by Taplin and associates, but undetectable in patients in complete remission after androgen ablation. This is consistent with measurement of expression of the androgen receptor using immunohistocytochemistry in androgen independent disease [56], and suggests that downregulation of the gene is not a frequent mechanism of androgen independence.

Amplification of the androgen receptor

Visakorpi and associates [57] have demonstrated that overamplification of the androgen receptor gene may occur in prostate carcinoma cells after androgen withdrawal, potentially making them more sensitive to lower concentrations of androgen.

Mutations in oncogenes or tumour suppressor genes to bypass androgen requirement

Various experiments on prostate cancer cell lines, and examination of tissue from human prostate cancers have shown that tumour cells may become androgen insensitive by mechanisms that do not involve the androgen receptor. Activation of proto-oncogenes, or inactivation of tumour suppressor genes, may allow the cell to proliferate in the absence of androgen, and to avoid apoptosis. For example, introduction of an activated *RAS* oncogene into the LNCaP cell line converts it to androgen independence [58], and other androgen-independent prostate cell lines have mutations in *RB1* tumour suppressor genes [59]. *BCL-2* is known to block cells from undergoing apoptosis, and its overexpression has been shown to be associated with the emergence of androgen independence in prostate cancer [60]. In addition, the *TP53* gene, which is also involved in the control of the cell cycle and of apoptosis is mutated in some hormone independent prostate cancers [61].

OPTIMAL TIMING OF HORMONAL TREATMENT

There is a debate as to the optimal timing of hormonal therapy in those patients with asymptomatic nodal and bony metastatic disease, and in those at high risk of having metastatic disease; that is stage T3 tumours with extracapsular spread. The molecular mechanisms and gene mutations underlying hormone resistance have been discussed above. It is likely that such mutations are present in a subgroup of tumour cells in the primary cancer, which is biologically heterogeneous and genetically unstable, but become more frequently expressed in metastases, or advanced disease. Cells with such mutations may become the more dominant cell type due to clonal selection, whilst non-mutant cells regress in response to androgen ablation. In this case, it might be advantageous to use androgen ablation early in the disease, while the number of hormone resistant cells is small, combined with treatments more effective against hormone resistant cells [62]. In addition, total androgen ablation would make the selection of a clone sensitive to lower concentrations of androgen less likely. An alternative theory is that as most cancers progress, the frequency of mutations increases, so androgen ablation may encourage the development of androgen receptor gene mutations as part of an adaptive process. In this case, it would be better to withhold androgen ablation until symptoms occur. There is experimental evidence that supports the clonal selection model, and intrinsic heterogeneity of the original tumour [63].

Patients with T3 and metastatic tumours were investigated by the Veterans Administration Cooperative Urological Research Group (U.S.A.), comparing DES therapy at diagnosis with placebo, although the placebo arm received hormonal treatment at progression [64]. Delayed (placebo) rather than early hormonal therapy did not change survival over 5–6 years, but the risk of developing metastatic disease was reduced from 50% at 5 years to less than 20% at 8 years in the DES treated group. The data from this study were re-evaluated in 1988 [65] with the conclusion that younger patients with high-grade metastatic disease did derive a survival benefit from early treatment. Since then, several non-randomised studies have suggested a benefit from early hormonal therapy, although these data may reflect selection bias [66, 67]. The results of an MRC randomised trial are awaited.

TOTAL ANDROGEN BLOCKADE

Since the adrenals secrete 5% of circulating testosterone, and sine 5α-dihydrotestosterone is also formed in the prostate gland from circulating adrenal precursors, the concept of total androgen blockade has developed. This combines orchidectomy or an LHRH agonist with an anti-androgen such as flutamide to counteract the stimulatory effects of 5α-dihydrotestosterone on prostate cancer cells. The evidence about the ability of adrenal androgens to maintain growth in the normal prostate and in prostatic cancer is conflicting. Studies on an androgen dependent tumour model, the Shionogi carcinoma, demonstrate androgen sensitivity at low androgen concentrations [68]. However, studies in men soon after castration or with hypogonadotrophic hypogonadism suggest the adrenal glands produce insufficient androgen to maintain proliferation and normal function of the prostate [69]. Clinical data in patients with metastatic prostate cancer who relapse after castration have a 30% response rate to adrenal androgen blockade, suggesting that 5α-dihydrotestosterone levels in their prostates are sufficient to maintain tumour growth.

Clinical trials of total androgen blockade have also had conflicting results. Crawford and associates [70] compared leuprolide alone with leuprolide plus flutamide and found significant improvement in median time to progression and median survival of 2.6 and 7.3 months, respectively. Improvements were more marked in a subgroup with minimal disease and good performance status. In other trials with castration as the control, there was no benefit to total androgen blockade [71, 72]. Tumour flare in the LHRH agonist treatment arm has been suggested as a reason for the different conclusions of their trials. An EORTC trial of orchidectomy alone versus flutamide plus zoladex has now shown an improvement for total androgen blockade in time to first progression of 25 weeks [73]. Median overall survival was prolonged by 7.3 months. Again, patients with few bony metastases and good performance status seemed to benefit the most. A meta-analysis of 22 randomised trials has now been performed, comparing surgical or medical castration with total androgen blockade [74]. There was only a trend to benefit from total androgen blockade, with a 3.6% non-significant improvement in 5-year survival.

The cost of treatment and the 'side-effects' of reduced potency and libido must also be considered. One method of reducing both cost and side-effects was explored by Goldenberg and associates [75], who used intermittent total androgen suppression for at least 6 months, then withheld treatment until the PSA rose again to between 10 and 20 ng/ml, when further treatment was begun. They reported a median time to progression of 2 years, with 40–45% of time off therapy. This group has previously argued a theoretical advantage for this approach. Studies on progression of the androgen-dependent Shionogi carcinoma showed that androgen withdrawal altered the ratio of stem cells in the tumour cell population. Numbers of stem cells were initially reduced, but at progression there was a 500-fold increase in androgen-independent stem cells. They argue that if androgens are replaced before progression begins, the surviving stem cells would produce an androgen-dependent tumour amenable to re-treatment by androgen withdrawal [76]. Obviously, a randomised study would be required to determine whether intermittent treatment has a greater or lesser effect on survival, compared to continuous total androgen blockade, and to conventional castration.

NEO-ADJUVANT HORMONE THERAPY

The use of neo-adjuvant androgen deprivation therapy is based on the hypothesis that reduction in the tumour volume prior to radiotherapy would lead to increased control of the primary tumour, without increasing total dose of radiation, as there would be fewer clonogenic cells to kill. Reduction in the radiation field size would also be expected to reduce morbidity from normal tissue damage. In addition, using hormonal treatment at diagnosis of apparently localised disease means there are likely to be a small proportion of androgen resistant cells in the tumour, with a higher chance of complete response. There are also theoretical disadvantages, for example, if dividing cancer cells go into G0 as a result of androgen ablation, they may be relatively more radioresistant. A reduction in visible prostate volume may still leave viable cancer cells outside the reduced volume, especially when there was extracapsular extension prior to the onset of hormonal therapy. Reducing radiation field size may then lead to a geographical miss.

A phase III randomised trial of total androgen ablation, with goserelin and flutamide, for 2 months before and during radiotherapy, compared with radiotherapy alone, in locally advanced disease has been performed by the RTOG [77]. This study demonstrated a reduction in local progression from 71% to 46% at 5 years, an improvement in progression-free survival from 15% to 36% at 5 years, but no change in overall survival, although this may require longer follow-up to demonstrate.

There are similar theoretical advantages to neo-adjuvant hormone therapy prior to surgery, including downstaging, downgrading and downsizing, making surgery easier, reducing the risk of positive resection margins, and reducing the risk of impotence and incontinence. In a pilot study, Fair and associates [78] demonstrated a marked fall in PSA in all patients, but inconclusive evidence of tumour downstaging, although some patients had pathologically complete tumour regression.

SECOND-LINE HORMONE THERAPY

The results of treatment with second-line hormones are disappointing. Response rates are of the order of 20–30% with a median duration of response of approximately 3–4 months. Median survival from progression on first-line agents is around 12 months. Second-line treatment usually consists of blockade of adrenal androgens with hydrocortisone, aminoglutethimide or ketoconazole, or the addition of anti-androgenic drugs to orchidectomy or LHRH agonists. Other investigational agents, such as suramin and aromatase inhibitors, have been discussed above. In patients who have been treated with flutamide, a small proportion may have a flutamide withdrawal response [48].

CONCLUSION

Understanding the physiology of androgen production, its role in the normal prostate and in prostate cancer, is vital in order to understand and predict the response of patients to hormonal manipulation. More insights are being gained into these complex interactions, some of which may lead to novel therapeutic manoeuvres. It was hoped that total androgen blockade would improve survival, despite con-

siderable cost and significant side-effects, but the recent evidence from a systematic overview does not support this early optimism. The case for neo-adjuvant hormone treatment appears theoretically attractive, but is not yet proven. Treatment of hormone resistant disease remains difficult and unsatisfactory, so preventing the emergence of hormone resistance, or new ways of treating resistant cells are important areas for continued research.

1. Cunha GR, Donjacour AA, Cooke PJ, *et al.* The endocrinology and developmental biology of the prostate. *Endocr Rev* 1987, **8**, 338–362.
2. McConnell JD. Physiologic basis of endocrine therapy for prostatic cancer. *Urol Clin N Am* 1991, **18**, 1–12.
3. Harper ME, Pike A, Peeling WB, Griffith D. Steroids of adrenal origin metabolised by human prostatic tissue in vivo and in vitro. *J Endocrinol* 1974, **60**, 117–125.
4. Geller J, Albert JD, Nachtsheim DA, Loza D. Comparison of prostatic cancer tissue DHT levels at the time of relapse following orchiectomy or oestrogen therapy. *J Urol* 1984, **132**, 693–696.
5. Smith C, Assimos D, Lee C, Grayhack JT. Metabolic action of prolactin on the prostate: independent of androgen action. *Prostate* 1985, **6**, 49.
6. Sibley PEC, Harper ME, Pike A, Peeling WB, Griffith D. Growth hormone and prostatic tumours: localisation using a monoclonal human growth hormone antibody. *J Endocrinol* 1984, **103**, 311–315.
7. Maddy SQ, Chisholm GD, Hawkins RA, Habib HK. Localisation of epidermal growth factor receptors in the human prostate by biochemical and immunocytochemical methods. *J Endocrinol* 1987, **113**, 147–153.
8. Wilson EM, Smith EP. Growth factors in the prostate. *Prog Clin Biol Res* 1987, **239**, 205–233.
9. Carruba G, Leake RE, Rinaldi F, *et al.* Steroid growth factor interactions in human prostate cancer cells. *Steroids* 1994, **59**, 412–420.
10. Conover CA, Perry JE, Tindall DJ. Endogenous cathepsin-D mediated hydrolysis of IGF-binding proteins in cultured human prostatic carcinoma cells. *J Clin Endocrinol Metab* 1995, **80**, 987–993.
11. di-Sant'Agnese PA. Neuroendocrine differentiation in human prostatic carcinoma. *Hum Pathol* 1992, **23**, 287–296.
12. Brinkman AO, Faber PW, van Rooj HCJ, *et al.* The human androgen receptor; domain structure, genomic organisation and regulation of expression. *J Steroid Biochem* 1989, **34**, 307–310.
13. Furuya Y, Lin XS, Walsh JC, Nelson WG, Isaacs JT. Androgen ablation induced programmed death of prostatic glandular cells does not involve recruitment into a defective cell cycle or p53 induction. *Endocrinology* 1995, **136**, 1898–1906.
14. Kypriana N, English HF, Isaacs JT. Programmed cell death during regression of PC-82 human prostate cancer following androgen ablation. *Cancer Res* 1990, **50**, 3748.
15. Sinha BK, Yamazaki H, Eliot HM, *et al.* Relationships between proto-oncogene expression and apoptosis induced by anticancer drugs in human prostate tumour cells. *Biochem Biophys Acta* 1995, **25**, 1970 12–18.
16. Huggins C, Hodges CV. The effect of castration, of oestrogen and of androgen injection on serum phosphatases in metastatic carcinoma of the prostate. *Cancer Res* 1941, **1**, 942.
17. Byar DP, Corle DK. Hormone therapy for prostate cancer: results of the VACURG studies. In Wittes RE, ed. Consensus development conference on the management of clinically localized prostate cancer. NCI Monograph No. 7, Washington, D.C., Government Printing Office, 1988, 165–170.
18. Parmar H, Phillips RH, Lightman SL, *et al.* Randomised controlled study of orchidectomy vs long acting D-trp-6-LHRH microcapsules in advanced prostatic carcinoma. *Lancet* 1985, **2**, 1201–1205.
19. Grayhack JT, Keeler TC, Kozlowski JM. Carcinoma of the prostate: hormonal therapy. *Cancer* 1987, **60**(Suppl.), 589–601.
20. Tomic R, Bergman G. Hormonal effects of cessation of oestrogen treatment for prostatic carcinoma. *J Urol* 1987, **138**, 801–803.
21. Schnokowski M, Dousete A, Ekman P, *et al.* Characterisation and measurement of the androgen receptor in human benign prostatic hyperplasia and prostatic carcinoma. *J Clin Endocrinol Metab* 1977, **45**, 920–930.
22. Hedlund PO, Gustafsson H, Sjogren S. Cardiovascular complications to treatment of prostatic cancer with estramustine phosphate or conventional oestrogen: a follow up of 212 randomised patients. *Scand J Urol Nephrol* 1980, **55**(Suppl.), 103–105.
23. The Leuprolide Study Group. Leuprolide vs diethylstilboestrol for metastatic prostatic cancer. *N Engl J Med* 1984, **311**, 1281–1286.
24. Pavone-Macaluso M, De Voogt HJ, Viggiano G, *et al.* Comparison of diethylstilboestrol, cyproterone acetate and medroxyprogesterone acetate in the treatment of advanced prostatic cancer. *J Urol* 1986, **136**, 624–630.
25. Committee on Safety of Medicines. Hepatic reactions with cyproterone acetate. *Curr Problems Pharmacovigilance* 1995, **21**, 1.
26. Sogani PC, Whitmore WF Jr. Experience with flutamide in previously untreated patients with advanced prostatic cancer. *J Urol* 1979, **122**, 640–643.
27. Decensi AU, Boccardo F, Guarneri D, *et al.* Monotherapy with nilutamide, a pure non-steroidal antiandrogen, in untreated patients with metastatic carcinoma of the prostate. *J Urol* 1991, **146**, 377–381.
28. Kennealey GT, Furr BJA. Use of the nonsteroidal anti-androgen casodex in advanced prostatic carcinoma. *Urol Clin N Am* 1991, **18**, 99–110.
29. Van den Bossche H, Lauwers W, Willensens G, Cools W. Ketoconazole: an inhibitor of the cytochrome p450 dependent testosterone biosynthesis. *Prog Clin Biol Res* 1985, **185A**, 187–196.
30. Santen RJ, Van den Borssche H, Symoens J, *et al.* Sites of action of low dose ketoconazole on androgen biosynthesis in men. *J Clin Endocrinol Metab* 1983, **57**, 732–736.
31. Sonimo N. The use of ketoconazole as an inhibitor of steroid production. *N Engl J Med* 1987, **317**, 813–818.
32. Trachtenberg J, Pont A. Ketoconazole therapy for advanced prostate cancer. *Lancet* 1984, **2**, 433–435.
33. Dowsett M, Shearer RJ, Ponder BAJ, Malone P. The effects of aminoglutethemide and hydrocortisone, alone and combined, on androgen levels in post-orchiectomy prostatic cancer patients. *Br J Cancer* 1988, **57**, 190.
34. Robinson MRG. Aminoglutethemide: medical adrenalectomy in the management of carcinoma of the prostate. A review after 6 years. *Br J Urol* 1980, **52**, 328.
35. Davies JH, Dowsett M, Jacobs S, *et al.* Aromatase inhibition: 4-hydroxyandrostenedione in advanced prostatic cancer. *Br J Cancer* 1992, **66**, 139–142.
36. Plowman PN, Perry LA, Chard T. Androgen suppression by hydrocortisone without aminoglutethimide in orchidectomised men with carcinoma of the prostate. *Br J Urol* 1987, **59**, 225–231.
37. Rittmaster RS. Finasteride. *N Engl J Med* 1994, **330**, 120–125.
38. Presti JC Jr, Fair WR, Andriole G, *et al.* Multicentre, randomised, double-blind, placebo controlled study to investigate the effect of finasteride (MK-906) on stage D prostate cancer. *J Urol* 1992, **148**, 1201–1204.
39. Myers C, Cooper M, Stern C, *et al.* Suramin—a novel growth factor antagonist with activity in hormone-refractory metastatic prostate cancer. *J Clin Oncol* 1992, **10**, 881–889.
40. Eisenberger MA, Reyno LM, Jodrell DI, *et al.* Suramin, an active drug for prostate cancer: interim observations in a phase I trial. *J Natl Cancer Inst* 1993, **85**, 611–621.
41. Pinski J, Reile H, Halmos G, Groot K, Schally AV. Inhibitory effects of somatostatin analogue RC-160 and bombesin/gastrin-releasing peptide antagonist RC-3095 on the growth of the androgen-independent Dunning R-3327-AT-1 rat prostate cancer. *Cancer Res* 1994, **54**, 169–174.
42. Bogden AE, Taylor JE, Moreau JP, Coy DH. Treatment of R3327 prostate tumours with a somatostatin analogue (somatulin) as adjuvant therapy following surgical castration. *Cancer Res* 1990, **50**, 2646–2650.

43. Maulard C, Richaud P, Droz JP, *et al.* Phase I–II study of the somatostatin analogue lanreotide in hormone-refractory prostate cancer. *Cancer Chemother Pharmacol* 1995, **36**, 259–262.
44. Verhelst J, De Longueville M, Ongena P, Denis L, Mahler C. Octreotide in advanced prostatic cancer relapsing under hormonal treatment. *Acta Urol Belg* 1994, **62**, 83–88.
45. Logothetis CJ, Hossan EA, Smith JL. SMS 201-995 in the treatment of refractory prostatic carcinoma. *Anticancer Res* 1994, **14**, 2731–2734.
46. Kockler H, Culig Z, Hobisch A, Cato ACB, Bartsch G. Androgen receptor alterations in prostatic carcinoma. *The Prostate* 1994, **25**, 266–273.
47. Veldscholte J, Ris-Staples C, Kuiper GGJM, *et al.* A mutation in the ligand binding domain of the androgen receptor of human LNCaP cells affects steroid binding characteristics and response to anti-androgens. *Biochem Biophys Res Comm* 1990, **173**, 534–540.
48. Scher HI, Kelly WK. Flutamide withdrawal syndrome: its impact on clinical trials in hormone-refractory prostate cancer. *J Clin Oncol* 1993, **11**, 1566–1572.
49. Newmark JR, Hardy DO, Tomb DC, *et al.* Androgen receptor gene mutations in human prostate cancer. *Proc Natl Acad Sci USA* 1992, **89**, 6319–6323.
50. Gaddipati JP, Mcleod DG, Heidenberg HB, *et al.* Frequent detection of codon 877 mutation in the androgen receptor gene in advanced prostate cancers. *Cancer Res* 1994, **54**, 2861–2864.
51. Culig Z, Hobisch A, Cronaeur MV, *et al.* Mutant androgen receptor detected in an advanced stage prostatic cancer is activated by adrenal androgens and progesterone. *Mol Endocrinol* 1993, **7**, 1541–1550.
52. Suzuki H, Sato N, Watabe Y, Masai M, Seini S, Shimazaki J. Androgen receptor gene mutation in human prostate cancer. *J Steroid Biochem Mol Biol* 1993, **46**, 759–765.
53. Tilley WD, Buchanan G, Hickey TE, Horsfall DJ. Detection of androgen receptor gene mutations in human prostate cancers by immunohistocytochemistry and PCR-SSCP. *Proc 86th Ann Meeting, Am Assoc Cancer Res*, Toronto, March 1995, abstract.
54. Taplin ME, Bubley GJ, Shuster TD, *et al.* Mutation of the androgen receptor gene in metastatic androgen-independent prostate cancer. *N Engl J Med* 1995, **532**, 1393–1398.
55. Tilley WD, Wilson CM, Marcelli M, McPhaul MJ. Androgen receptor gene expression in human prostate cancer cell lines. *Cancer Res* 1990, **50**, 5382–5386.
56. Puizeveld de Winter JA, Janssen PJA, Sleddens HMEB, *et al.* Androgen receptor status in localised and locally progressive hormone-refractory human prostate cancer. *Am J Pathol* 1994, **144**, 735–736.
57. Visakorpi T, Hyytinen E, Kovisto P, *et al.* Amplification of the androgen receptor gene is common in recurrent prostate cancer from patients treated with androgen withdrawal. *Proc Am Urol Assoc*, Las Vegas, April 1995, **153**(Suppl 4), 379A (abstract).
58. Voeller HJ, Wildding G, Gelmann EP. v-H-ras expression confers hormone independent growth to LNCaP prostate cancer cells. *Mol Endocrinol* 1991, **5**, 209–216.
59. Rubin SJ, Hallahan DE, Ashman CR, *et al.* Two prostate cancer cell lines demonstrate abnormalities in tumour suppressor genes. *J Surg Oncol* 1991, **46**, 31–36.
60. McDonnell TJ, Troncoso P, Brishay SM, *et al.* Expression of the proto-oncogene bcl-2 in the prostate and its association with emergence of androgen-independent prostate cancer. *Cancer Res* 1992, **52**, 6940–6944.
61. Bookstein R, MacGrogen D, Hilsenbeck SG, Sharkey F, Allieo DC. p53 is mutated in a subset of advanced stage prostate cancer. *Cancer Res* 1993, **53**, 3369–3373.
62. Kozlowski JM, Ellis WJ, Grayhack JT. Advanced prostatic carcinoma. Early versus late endocrine therapy. *Urol Clin N Am* 1991, **18**, 15–25.
63. Carter HB, Isaacs JT. Experimental and theoretical basis for hormonal treatment of prostatic cancer. *J Urol* 1988, **140**, 173.
64. Byar DP. The Veterans Administration Cooperative Urological Research Group's studies of cancer of the prostate. *Cancer* 1973, **32**, 1126.
65. Byar DP, Coyle D. Hormone therapy for prostate cancer: results of the Veterans Administration Cooperative Urological Research Group's studies. *Natl Cancer Inst Monogr* 1988, **7**, 165.
66. Va Aubel OG, Hoelistra WJ, Schroder FH. Early orchiectomy for patients with stage D1 prostatic carcinoma. *J Urol* 1985, **134**, 292.
67. Zinder H. Extended experience with surgical treatment of stage D1 adenocarcinoma of the prostate. Significant influences of immediate adjuvant hormonal treatment on outcome. *J Urol* 1989, **23**(Suppl. 5), 27.
68. Labrie F, Vailleux F, Fournier A. Low androgen levels induce the development of androgen hypersensitive cell clones in Shionogi mouse mammary carcinoma cells in culture. *J Natl Cancer Inst* 1988, **80**, 1138–1147.
69. Oesterling JE, Epstein JI, Walsh PC. The viability of adrenal androgens to stimulate the adult human prostate—an autopsy evaluation of men with hypogonadotrophic hypogonadism and panhypopituitarism. *J Urol* 1986, **136**, 1030–1034.
70. Crawford ED, Eisenberger MA, Mcleod DG, *et al.* A controlled trial of leuprolide with and without flutamide in prostatic carcinoma. *N Engl J Med* 1989, **321**, 419–424.
71. Iverson P, Sucia S, Sylvester R, Christensen I, Denis L. Zoladex and flutamide versus orchidectomy in the treatment of advanced prostatic cancer. *Cancer* 1990, **66**, 1067–1073.
72. Beland G, Elhilali M, Fradet Y, *et al.* A controlled trial of castration with and without nilutamide in metastatic prostatic carcinoma. *Cancer* 1990, **66**, 1074–1079.
73. Denis LJ, Carneiro de Moura JL, Bono A, *et al.* Goserelin acetate and flutamide versus bilateral orchidectomy: a phase III EORTC trial. *Urology* 1993, **42**, 119–130.
74. Prostate Cancer Triallists' Collaborative Group. Maximum androgen blockade in advanced prostate cancer: an overview of 22 randomised trials with 3283 deaths in 5710 patients. *Lancet* 1995, **346**, 265–269.
75. Goldenberg SL, Bruchovsky N, Gleave ME, Sullivan LD, Akakura K. Intermittent androgen suppression in the treatment of prostate cancer: a preliminary report. *Urology* 1995, **45**, 839–844.
76. Akakura K, Bruchovsky N, Goldenberg SL, Rennie PS, Buckly AR, Sullivan LD. Effects of intermittent androgen suppression on androgen dependent tumours. *Cancer* 1993, **71**, 2782–2790.
77. Pilepich MV, Sause WT, Shipley WU, *et al.* Androgen deprivation with radiation therapy compared with radiation therapy alone for locally advanced prostatic carcinoma: a randomised comparative trial of the radiation therapy oncology group. *Urology* 1995, **45**, 616–623.
78. Fair WR, Aprikian A, Sogani P, Reuter V, Whitmore WF. The role of neoadjuvant hormonal manipulation in localised prostatic cancer. *Cancer* 1993, **71**, 1031–1038.