LEADING ARTICLE

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Testosterone

Its Role in Development of Prostate Cancer and Potential Risk From Use as Hormone Replacement Therapy

Sarah Slater and *R.T.D. Oliver*

Department of Medical Oncology, St Bartholomew's and The Royal London Hospital School of Medicine, West Smithfield, London, England

Abstract

Evidence from studies in patients with prostate cancer of intermittent hormone therapy combined with results from rechallenge of hormone resistant patients with testosterone demonstrate that the majority of prostate cancers retain a similar degree of dependence on male sex hormone milieu as normal prostate cells. Yet there has so far been no conclusive evidence, despite 34 studies, that levels of circulating testosterone in individuals developing prostate cancer are higher than in controls. The aim of this article was to critically evaluate this evidence and seek clues to other mechanisms whereby sex hormones could influence the development of prostate cancer. Additionally, epidemiological data were examined to investigate the interplay between sex hormone levels and environmental factors to help understand the development of prostate cancer and identify a safe way to provide hormone replacement therapy (HRT).

Three overviews provide similar evidence that there is no significant difference in mean testosterone levels between patients and controls. However in the most recent review of studies, though there was no difference in means between cases and controls, there was a significant risk (adjusted odds ratio 2.34) for individuals identified by comparing incidence of prostate cancer in men in the upper and lower quartile of testosterone level. This report, taken with epidemiological data demonstrating that prostate cancer risk is increased by early age of onset of sexual activity and multiple nonspecific sexually transmitted diseases (STDs), has led to the hypothesis that the link between sex hormones and prostate cancer is indirect. Those individuals with high testosterone levels were more at risk of acquisition of multiple nonspecific STDs. This promotes transformation of prostate cells and damage to Leydig cells in the testis leading to there being no difference in testosterone compared with controls by the time the tumour is diagnosed.

Because of the observed relationship between testosterone and prostate cancer development there has been anxiety about marketing HRT for men. Two observations support the view that the prostate cancer risks from use of testosterone hormone replacement may not be as great as first feared. Firstly, prostate cancers arising in men with low serum testosterone levels are more malignant and frequently nonresponsive to hormones. Secondly, breast cancers diagnosed in women on HRT though increased in number are less malignant possibly because of enhanced sensitivity to hormone therapy, and the situation may prove to be analogous with prostate cancer and testosterone replacement.

That prostate cancer can be regulated by sex hormone milieu is beyond doubt, based on the pioneering experiments of Huggins and Hodges[1] on castration and stilboestrol that have now been translated into the standard treatment for patients with metastatic disease.

In doubt, however, is the issue of whether this effect is mediated by testosterone itself or whether it is a change in other hormones, induced by surgical or chemical castration. The reason for this is conflicting findings from recent reviews of studies that have attempted to investigate the effects of serum testosterone levels on prostate cancer development. In the first review, 4 studies showed that high serum testosterone levels were associated with an elevated risk of prostate cancer, 6 studies demonstrated there was a reduced risk and 15 showed no difference compared with a control population.[2] The second overview, which was of prospective studies only, included 2 from the previous overview in addition to 6 more recent studies. However, these studies still failed to demonstrate a significant association.[3] In contrast the third review found a highly significant increase in risk [odds ratio (OR) 2.3] but only when analysing those patients with serum hormone levels in the upper quartile.[4] Resolving this issue is of considerable importance in terms of clarifying the risk of testosterone hormone replacement therapy (HRT) in the development of prostate cancer in men. Development of reliable HRT preparations for men is much less advanced than in women, in part because of the fear of a medicolegal suit should any product be shown to be associated with an increased risk of prostate cancer. As it could take more than 5 to 10 years for any increase of risk to appear progress is considerably slow in this area.

This paper sets out to clarify the reasons for the failure to clearly define the risk posed by testosterone in the development of prostate cancer by reviewing: (i) current knowledge about endocrinology of the normal prostate; (ii) age-related changes in sex hormone levels; (iii) current understanding of the epidemiology of prostate cancer, before undertaking a further systematic review of the literature that has been published on the issue of serum testosterone and prostate cancer.

1. Physiology of Sex Hormone Control of the Normal Prostate

The first step in unravelling the conflicting evidence lies in understanding the complex set of pathways which link the hypothalamic-pituitary axis to the adrenal glands, testes and prostate, and lead to androgen production. Gonadotrophin-releasing hormone, is secreted in pulses from the hypothalamus into the pituitary gland, causing the subsequent pulse release of luteinising hormone (LH) and follicle-stimulating hormone (FSH). LH stimulates the testicular Leydig cells to produce the androgens, testosterone and dihydrotestosterone (DHT), via the enzyme 5α-reductase, and oestradiol. FSH along with testosterone stimulates the testicular Sertoli cells to release the hormone inhibin and induces sperm production.[5]

Androgens are essential for the normal growth and maintenance of the prostate gland. Apoptosis is activated in both normal prostate and cancer cells when androgens are withdrawn.[6,7] The principal circulating androgen in serum is testosterone, the majority of which is bound to sex hormone-binding globulin (SHBG) and thus is inactive. SHBG is a β-globulin with a molecular weight of 94kD, which is synthesised in the liver and has one binding site per mole. It transports both testosterone and oestradiol, but has lower affinity for oestradiol. The median serum concentration of this protein in men is 32 μmol/L and in women is 1.5 times this level.[8] SHBG levels fall in the second decade of life, irrespective of androgen status. They are increased by estrogen therapy, androgen deficiency, hyperthyroidism, acute severe illness and hepatic cirrhosis.

In the prostate, testosterone is converted into the more potent form, DHT, by 5α-reductase isoenzymes. DHT is essential for the development of normal external genitalia in the male foetus.[9] It binds to the intracellular androgen receptor (AR), and the resultant complex acts as the main regulator of cell proliferation and secretory function of the

prostate. In addition, testosterone is converted into oestradiol in adipose tissue, the liver and in the central nervous system.

Testosterone function is genetically controlled by 4 groups of genes.[10-14] The first is the AR gene itself, which is polymorphic and its protein product varies in its affinity for testosterone due to the variable number of microsatellite regions within the gene. The frequency of these variants differs in different races;[10] this could contribute to differences in the lifetime risk for the effects of a given dose of testosterone. Abnormalities in the allele frequencies of microsatellites at the AR locus also differ significantly amongst healthy African-Americans, Caucasians and Asian men. African-Americans have a high prevalence of short CAG microsatellite alleles and low frequency of 16 GGC repeats compared with Caucasians and Asians. The belief is that shorter CAG microsatellite alleles of the AR gene cause more active growth in the prostate cells, increasing the potential risk of prostate cancer.[10-14]

The second, more recently described, method of genetic control of testosterone function is via the cytochrome (CYP) CYP3A4 gene which is involved in hydroxylation of testosterone. As this is the first step in its deactivation, low levels of the gene product would slow the rate of inactivation and increase the biological effect of a given serum level of testosterone.[11] Rebbeck et al.[11] evaluated the role of CYP3A4 in 230 patients with cancer of the prostate. They found that of the 34 patients who had a novel mutation upstream of this gene, 41% had locally advanced tumours compared with 24% of patients without this variant ($p = 0.049$). Their hypothesis proposed that the mutation associated with the CYP3A4 gene was responsible for the clinical presentation of prostate cancer; this is supported by the fact that CYP3A4 is responsible for the hydroxylation of testosterone, therefore a mutation in this gene may reduce the functional deactivation of the hormone. Thus, men with the variant genotype may have decreased protein activity and therefore decreased testosterone hydroxylation. This may in turn lead to an increase in the bioavailability of testosterone for conversion to its intracellular mediator, DHT.[11]

The third mechanism is via a gene affecting conversion of testosterone to the active intraprostatic androgen DHT, with linkage of increased risk of prostate cancer to alleles that increase the efficiency of the 5α-reductase gene.[12] The fourth mechanism of control occurs via the expression of a genetic variant which leads to elevated levels of DHT and thus, potentially, a predilection to prostate cancer.[13]

Though it is accepted that levels of testosterone decline with age, these differences are not great. However, the fact that SHBG levels rise with age provides evidence that the testosterone levels in older men are significantly lower. SHBG levels are higher in patients with prostate cancer who are over the age of 70 years, compared with younger men ($p = 0.02$).[2] It is noteworthy that SHBG levels also have a tendency to be higher in men with a healthy prostate who are over 70 years old.[2] Racial differences are also evident; compared with Caucasians, Blacks have SHBG levels which are 10% higher. Levels of SHBG are higher in both Blacks and Caucasians compared with Japanese men, by 53 and 10%, respectively ($p < 0.01$).[15]

The precise role of the AR in prostate cancer is in itself unclear. The receptor is found predominantly in the nuclei of glandular epithelial cells in a normal prostate, benign prostatic hyperplasia and prostatic cancer. In the latter, there are both AR-positive and -negative cells. There does not seem however, to be any correlation between the percentage of positive cells and time to disease progression after androgen ablation.[15] In hormone-refractory cancer, point mutations in the receptor have been identified, which increase in number as the cancer becomes hormone resistant.[16-20]

2. Aetiological Factors in Prostate Cancer Development

Prostate cancer is primarily a disease of the elderly with the majority of deaths occurring in patients over the age of 65 years.[5] It has long been known from postmortem studies that early-

stage cancer can be detected long before this, and up to 70% of elderly men may demonstrate histological changes of prostate cancer without dying from it.[21] Of greater interest is the fact that the rate of death from cancer of the prostate is highest in African-Americans, while Japanese males have one of the lowest rates in the world. However, the frequency of latent cancers defined at postmortem is the same in all 3 racial groups.[21] More recently, the classification of histological changes of prostatic intra-epithelial neoplasia has improved and postmortem studies are demonstrating that these changes may be present in up to 30% of 40-year-old patients with equal prevalence in all racial groups.[22]

In terms of aetiological risk factors there are 4 main classes, sexual, dietary, physico-chemical and genetic; sexual and dietary risk factors are outlined in table I.[23] Patients with prostate cancer have on average an earlier onset of sexual activity and an increased frequency of low grade nonspecific prostatitis possibly induced by chlamydia.[24] This association has led to the suggestion that low grade prostatitis acts as a promotional factor involved in converting latent to clinically relevant cancer.[25] Dietary factors include a high intake of animal fat, low intake of vitamin A-related compounds such as lycopenes, low levels of vitamin D and low sun exposure.[23,24] As vitamin A deficiency reduces T cell immune responses and vitamin D deficiency reduces macrophage functions, a possible mechanism which could link these potential risk factors is that they suppress the immune response and could potentially increase the persistence of 'nonspecific prostatitis' in patients with this disease.[25]

Physico-chemical exposures associated with prostate cancer development include radiation in nuclear plants, the displacement of zinc by cadmium because the prostate has high avidity for heavy metals for the regulation of citric acid synthesis. The final chemical factor is exposure to agricultural chemicals such as organo-chlorine pesticides.[26,27]

The final risk factor is genetics. Prostate cancer is clearly increased in families with a history of this disease and a major effort is in progress to identify the prostate cancer gene. It is clear that there are likely to be several genes involved, because as well as an association with the breast cancer associated (BRCA)1/2 group of genes there is an association with chromosome 1 and evidence of a tumour suppressor gene on chromosome 10.[28,29] As yet, none of these genetic associations have been linked to a testosterone-mediated mechanism, though as outlined in section 3 there are possible genetic mechanisms involved.

Table I. Overview of dietary and sexual behaviour risk factors for prostate cancer (adapted from Key[23])

Risk factors	Relative risk
Obesity	1.25
Meat eater	1.34
High dairy product consumption	1.30
High total fat intake	1.31
High animal fat intake	1.54
High carrot consumption	0.66
High green vegetable consumption	0.93
Early age of first intercourse	1.31
High numbers of sexual partners	1.21
Sexually acquired infection	1.86
Syphilis	0.77
Gonorrhoea	1.22
Vasectomy	1.54

3. Evidence for Involvement of Testosterone in Prostate Cancer Development

That sex hormones are involved in the regulation of the prostate and in the aetiology of prostate cancer is beyond dispute. The first article on the dependence of the normal prostate gland and its malignancies on hormonal function was published by John Hunter in 1785.[30] Less than 200 years later in 1941 Huggins and Hodges re-emphasised the observation that castration by the administration of estrogens caused regression of advanced prostate cancer in dogs and led to the widespread use of this approach clinically.[1] Though this observation is critical in making the case for an aetiological role for androgens and testosterone in pros-

Table II. 5α-Reductase metabolites in Japanese compared with Caucasian and African American young postpubertal men in the US (modified from Ross et al.[32])

Metabolites	Japanese (n = 54)	Caucasian (n = 47)	African American (n = 49)
Androsterone (ng/ml)	49	73	69
3α, 17β androstanediol (ng/ml)	5.3	6.9	6.6
Testosterone (ng/dl)	602	575	640

tate cancer development, there are other associations that reinforce this view.

Firstly, the clinical incidence of prostate cancer varies significantly across the world, with the highest incidence occurring in African-Americans (79 per 100 000) and the lowest in Japanese males (4 per 100 000).[5] Ross et al.[31] have demonstrated that at the time of puberty African American males have 10 to 15% higher levels of circulating testosterone than their Caucasian counterparts, but equal levels compared with Japanese men (table II), who because of a genetic deficiency of 5α-reductase actually have lower DHT levels in the prostate. In addition, differences in the function of 5α-reductase genes affecting the AR and androgen metabolism contribute to an increased risk of prostate cancer in African-American men.[10-14,33]

Secondly, prostate cancer can be induced in rats to whom large amounts of testosterone have been administered.[34,35] Thirdly, men castrated prior to puberty never develop prostate cancer.[1] A reduced risk of this cancer has been also been associated with hyperoestrogenic states (e.g. cirrhosis cases),[36] and estrogen therapy has a palliative role in advanced prostate cancer because it competes with testosterone in the hypothalamus and suppresses gonadotropin production. Finally, prostate cancer may be successfully treated by surgical or medical androgen ablation.

On the basis of this evidence alone, the case that raised serum testosterone levels are a major factor in development of cancer of the prostate seems to be a logical presumption. However, there have been 30 clinical studies published over the past 32 years with conflicting results; these studies have been reviewed elsewhere.[2-4] The first 2 reviews used an analysis of mean values and found no differences in testosterone levels between patients with prostate cancer and those without (tables III and IV), while the most recent study analysed patients in the upper quartile for testosterone levels as compared with those in the lowest quartile and provided evidence of a significant association between serum testosterone levels and risk of prostate cancer (table V). The authors also highlighted the only clinical study to provide a multivariate analysis of adjusted SHBG data and showed that the SHBG levels have an inverse correlation with cancer risk (OR 0.46).[4] In addition, this review also highlighted the consistent association of increased risk with elevated insulin-like growth factor-1 in 3 of 3 studies. There is increasing evidence that this growth factor may play an autocrine stimulatory role in the progression of cancer. As yet there are no studies investigating interaction between testosterone and insulin-like growth factors.

3.1 Testosterone Effects on Active Prostate Cancer

There are 2 sources of information about the effect of testosterone on active prostate cancer. The first has emerged from studies using intermittent hormone therapy. Clinical trials were carried out

Table III. 1993 overview of unselected studies comparing testosterone levels in controls and patients with prostate cancer at time of diagnosis (modified from Andersson et al.[2])

Outcome of study	No. of studies	No. of participants in the studies	
		patients	controls
Total	25	1481	2767
Mean testosterone level higher in patients than controls	4	343	503
Mean testosterone level same in patients *vs* controls	15	758	2004
Mean testosterone level lower in patients than control	6	380	260

Table IV. 1999 review of prospective case control studies investigating differences in hormone levels between patients with prostate cancer and controls (modified from Shaneyfelt et al.[4])

Hormone	No. of patients	No. of controls	No. of studies	Ratio of mean hormone level between patients and controls (95% CI)
Testosterone	817	2107	8	0.99 (0.95-1.02)
Non-SHBG-bound testosterone	325	442	3	1.02 (0.96-1.09)
Dihydrotestosterone	636	1040	5	0.98 (0.94-1.03)
Androstanediol glucuronide	644	1048	5	1.05 (1.00-1.11)
Androstenedione	316	1320	3	1.01 (0.97-1.06)
Dehydroepiandrosterone	197	312	2	0.98 (0.88-1.10)

CI = confidence interval; **SHBG** = sex hormone-binding globulin.

after animal studies provided some evidence that intermittent re-exposure to testosterone decreases the rate of progression to hormone-resistant cancer.[37] In these studies patients received 6 to 12 months of hormone therapy and then remained off treatment for 9 to 12 months, with a recurrence of prostate specific antigen (PSA) tumour activity following closely after recovery of testosterone levels.[38] In the average patient, these cycles can be repeated 2 to 4 times before hormone resistance occurs, although the time to hormone resistance is double that of patients who are treated continuously.[39] In the majority, but not all, patients the speed of recovery of PSA levels in each cycle matches the recovery of testosterone production.

The second type of data comes from limited studies where patients with hormone-resistant cancer have been re-exposed to testosterone either on its own or to try to increase proliferation of tumour cells in order to enhance the response to subsequently given antimitotic chemotherapy. Though these studies failed to show a response to chemotherapy, the majority of patients had increased cancer activity during the period on testosterone, demonstrating that testosterone dependence persists even in hormone-resistant patients.[40]

Further evidence of the effect of testosterone levels on active prostate cancer comes from clinical studies correlating pretreatment testosterone with outcome. Initially conflicting observations have been made. Patients without metastases who demonstrated a higher level of testosterone before radiation also had a higher risk of relapse after radiation.[41] As radiation, presumably due to scatter to the testis, reduces testosterone levels after treatment,[42] those with high testosterone levels are less likely to be affected by radiation. Another observation is the correlation of pre-hormone therapy testosterone levels with outcome after androgen ablation. Two studies[43,44] have provided conclusive evidence that the higher the patient's pretreatment testosterone level, the better the chance of response to androgen withdrawal.

4. Discussion and Conclusion

From this review, it is clear from multiple population-based studies that the link between testosterone levels and the development of prostate cancer is at best weak and not as clear cut as one would have surmised at the outset. Despite this evidence, there is little dispute that testosterone

Table V. Unadjusted and adjusted odds ratio for prostate cancer for patients with sex hormone levels in the upper quartile compared with the lowest quartile in selected prospective studies (modified from Shaneyfelt et al.[4])

	Unadjusted odds ratio	Adjusted odds ratio[a]
Testosterone	1.25 (0.9-1.8)	2.34 (1.3-4.2)
Dihydrotestosterone	0.87 (0.6-1.3)	0.71 (0.4-1.3)
Sex hormone-binding globulin	0.69 (0.4-1.2)	0.46 (0.2-0.9)
Insulin-like growth factor-1	NA	1.91 (1.5-3.3)

a Adjusted for body mass and other steroid levels.

NA = not available.

suppression results in clinical remissions in patients with prostate cancer. However, the data from epidemiology studies are only conclusive when focusing on patients in the upper 25th percentile or studying racially homogenous groups for example, in the case of the deficient functioning of 5α-reductase in Japanese men.

The weakness of the overall effect of testosterone can be explained in 2 possible ways. One explanation is that testosterone plays no apparent role in the development of prostate cancer. Therefore the association with a response is due to an associated change such as altered LH or 5α-reductase function leading to a disparity between serum and intraprostatic levels and androgens. Alternatively, testosterone influence may be modified by other circumstances, such as different genetic predispositions, diets and infectious damage impacting at different stages of cancer development. With increasing numbers of genes now known to affect genetic androgen metabolism, the latter explanation seems more likely.

Available data suggest that there may be an increased susceptibility at the cellular level of either receptor function or hormone metabolism in prostate cancer patients.[32,45]

A final confounding issue relates to intra-uterine development. In early gestational studies, Black women have testosterone levels that are almost 50% higher than those in Caucasian women. This may lead to an altered steroid secretion or receptor sensitivity in their offspring.[46]

Given these provisos, it is beyond all reasonable doubt that testosterone does promote the growth of prostate cancer and that populations with high levels of intraprostatic DHT at the time of puberty are at an increased risk of prostate cancer. However, in later life when prostate cancer presents, its relationship to testosterone levels is less apparent. This raises the question as to whether the increased numbers of sexually acquired diseases that men with these high testosterone levels acquire in early puberty is damaging the testes and lowering testosterone production in later life.[47,48]

This could explain why there is not a clear picture relating the occurrence of prostate cancer to testosterone levels at the time of diagnosis. There may be 2 processes involved in the cancer progression, such as hormone-driven and infection-driven effects. The latter may be more dominant by the time the disease becomes clinically significant; it may also cause a low grade orchitis at the same time and damage hormone production by the testis, as has been demonstrated after mumps orchitis.[49,50] This in turn may lead to an apparent paradox of high testosterone in young men at risk of prostate cancer subsequently leading to lower levels at the time of presentation.

From a practical point of view, the conclusion from this overview is that testosterone is a factor, albeit indirect, acting via intraprostatic androgens in the development of prostate cancer. If latent prostate cancer is present, it is highly likely that administering testosterone HRT will increase the chances that a proportion of these cancers will progress to become clinically evident. Presumably such tumours, if they did develop, would retain hormone sensitivity that could have survival advantage as it is well established that prostate cancers arising in men with low testosterone levels have a poor prognosis for response to therapy.[44] There is an increasing amount of evidence from the study of breast cancer that a similar situation occurs in this tumour. There is increasing evidence that latent breast cancer exists as frequently as does latent prostate cancer. There is already some evidence that estrogen-based HRT increases the number of these latent breast cancers that present clinically. These tumours tend to be less malignant and have a higher frequency of good prognosis localised oestrogen receptor-positive tumours.[51,52] As a consequence there is no evidence that HRT is associated with any increase in breast cancer mortality. If testosterone HRT were to become more widely available for men, it is likely that some of the latent prostate cancers would become manifest clinically, though hopefully like the breast cancers in women on HRT, they would also be less malignant. It is also possible that testosterone HRT might

have other positive benefits in reducing the risk of prostate cancer. There is already some evidence from studies in women demonstrating that oxytocin release at the time of orgasm, by clearing breast ducts of cumulated carcinogens, may reduce the incidence of invasive breast cancer.[53] Given the high incidence of impotence in men presenting with clinically significant prostate cancer, it is possible that testosterone HRT possibly combined with sildenafil could also provide a similar benefit for men.[54,55]

References

1. Huggins C, Hodges C. Studies on prostatic cancer 1: the effect of castration, of estrogen and of androgen injection on serum phosphatases in metastatic carcinoma of the prostate. Cancer Res 1941; I: 293-7
2. Andersson SO, Adami HO, Bergstrom R, et al. Serum pituitary and sex steroid hormone levels in the etiology of prostatic cancer: a population-based case-control study. Br J Cancer 1993; 68 (1): 97-102
3. Eaton NE, Reeves GK, Appleby PN, et al. Endogenous sex hormones and prostate cancer: a quantitative review of prospective studies. Br J Cancer 1999; 80 (7): 930-4
4. Shaneyfelt T, Husein R, Bebkey G, Mantzoras C. Hormonal predictors of prostate cancer: a meta analysis. J Clin Oncol 2000; 18 (4): 847-53
5. Oesterling J, Fuks Z, Lee CT. Cancer of the prostate. In: Devita V, Hellman S, Rosenberg S, editors. Cancer: principles and practises in oncology. 5th ed. New York: Lippincott-Raven, 1997
6. Colombel M, Olsson CA, Ng PY, et al. Hormone-regulated apoptosis results from reentry of differentiated prostate cells onto a defective cell cycle. Cancer Res 1992; 52 (16): 4313-9
7. Kyprianou N, English HF, Isaacs JT. Programmed cell death during regression of PC-82 human prostate cancer following androgen ablation. Cancer Res 1990; 50 (12): 3748-53
8. Ahluwalia B, Jackson MA, Jones GW, et al. Blood hormone profiles in prostate cancer patients in high-risk and low-risk populations. Cancer 1981; 48 (10): 2267-73
9. Bruchovsky N, Wilson JD. The conversion of testosterone to 5-alpha-androstan-17-beta-ol-3-one by rat prostate *in vivo* and *in vitro*. J Biol Chem 1968; 243 (8): 2012-21
10. Giovannucci E, Stampfer M, Krithivas K, et al. The CAG repeat within the androgen receptor gene and its relationship to prostate cancer. Proc Natl Acad Sci U S A 1997; 94 (7): 3320-3
11. Rebbeck TR, Jaffe JM, Walker AH, et al. Modification of clinical presentation of prostate tumours by a novel genetic variant in CYP3A4. J Natl Cancer Inst 1998; 90 (16): 1225-9
12. Makridakis N, Ross R, Pike M, et al. Association of mis-sense substitution in SRD5A2 gene with prostate cancer in African-American and Hispanic men in Los Angeles, USA. Lancet 1999; 354 (9183): 975-8
13. Devgan SA, Henderson BE, Yu MC, et al. Genetic variation of 3 beta-hydroxysteroid dehydrogenase type II in three racial/ethnic groups: implications for prostate cancer risk. Prostate 1997; 33 (1): 9-12
14. Irvine RA, Yu MC, Ross RK, et al. The CAG and GGC microsatellites of the androgen receptor gene are in linkage disequilibrium in men with prostate cancer. Cancer Res 1995; 55 (9): 1937-40
15. Wilding G. Endocrine control of prostate cancer. Cancer Surv 1995; 23: 43-62
16. Suzuki H, Sato N, Watabe Y, et al. Androgen receptor gene mutations in human prostate cancer. J Steroid Biochem Mol Biol 1993; 46 (6): 759-65
17. Culig Z, Hobisch A, Cronauer MV, et al. Mutant androgen receptor detected in an advanced-stage prostatic carcinoma is activated by adrenal androgens and progesterone. Mol Endocrinol 1993; 7 (12): 1541-50
18. 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 (11): 2861-4
19. Newmark JR, Hardy DO, Tonb DC, et al. Androgen receptor gene mutations in human prostate cancer. Proc Natl Acad Sci U S A 1992; 89 (14): 6319-23
20. Scher HI, Zhang ZF, Nanus D, et al. Hormone and antihormone withdrawal: implications for the management of androgen-independent prostate cancer. Urology 1996; 47 (1A Suppl.): 61-9
21. Oishi K, Yoshida O, Schroeder F. The geography of prostate cancer and its treatment in Japan. Cancer Surv 1995; 23: 267-80
22. Sakr WA, Grignon DJ, Hass G, et al. Epidemiology of high grade intraepithhelial neoplasia. Pathol Res Pract 1995; 191: 838-41
23. Key T. Risk factors for prostate cancer. Cancer Surv 1995; 23: 63-77
24. Ostaszewska I, Stefano-Zdrodowska B, Badyda J, et al. Chlamydia trachomatis: probable cause of prostatitis. Int J STD AIDS 1998; 9 (6): 350-3
25. Oliver RTD. Adjuncts for magnifying the effectiveness of intermittent hormone therapy in early and advanced prostate cancer. In: Belldegrun A, Kirby RS, Oliver RTD, editors. New perspectives in prostate cancer. Oxford: ISIS Medical Media, 1998: 325-36
26. Dich J, Wiklund K. Prostate cancer in pesticide applicators in Swedish agriculture. Prostate 1998; 34 (2): 100-12
27. Sharma-Wagner S, Cookalingam A, Malker H. Occupation and prostate cancer risk in Sweden. J Occup Environ Med 2000; 42 (5): 517-25
28. Bishop D, Kiemeney L. Family studies and the evidence for genetic susceptibility to prostate cancer. Semin Cancer Biol 1997; 8 (1): 45-51
29. Ittmann M. Chromosome 10 alterations in prostate adenocarcinoma. Oncol Rep 1998; 5 (6): 1329-35
30. Hunter J. Observations on certain parts of the animal economy. Bibliotheca Osteriana. London: England, 1785
31. Ross R, Pike M, Coetzee G, et al. Androgen metabolism and prostate cancer: establishing a model of genetic susceptibility. Cancer Res 1998; 58 (20): 4497-504
32. Ross R, Bernstein L, Lobo R. 5-alpha-reductase activity and risk of prostate cancer among Japanese and US white and black males. Lancet 1992; 339: 887-9
33. Pettaway C. Racial differences in the androgen/androgen receptor pathway in prostate cancer. J Natl Med Assoc 1999; 91 (12): 6536-0
34. Henderson BE, Ross RK, Pike MC, eta l. Endogenous hormones as a major factor in human cancer. Cancer Res 1982; 42 (8): 3232-9

35. Nobel R. The development of prostatic adenocarcinoma in Nb rats, following prolonged sex hormone administration. Cancer Res 1977; 37: 1929-33
36. Glantz C. Cirrhosis and carcinoma of the prostate gland. J Urol 1964; 91: 291-3
37. Sato N, Gleave ME, Bruchovsky N. Intermittent androgen suppression delays progression to androgen-independent regulation of prostate-specific antigen gene in the LNCap prostate tumour model. J Steroid Biochem Mol Biol 1996; 58 (2): 139-46
38. Oliver RTD. Intermittent hormone therapy: its potential in early prostate cancer and intra-epithelial neoplasia. Jpn J Cancer Chemother 2000; 27 (2): 399-404
39. Oliver R, Grant-Williams G, Paris A, et al. Intermittent androgen deprivation after PSA complete response as a strategy to reduce induction of hormone resistant prostate cancer. Urology 1997; 49: 79-82
40. Fowler J, Whitmore W. The response of metastatic adenocarcinoma of the prostate to exogenous testosterone. J Urol 1981; 126 (3): 372-5
41. Zagars G, Pollack A, Eschenbach A. Serum testosterone: a significant determinant of metastatic relapse for irradiated localised prostate cancer. Urology 1997; 49: 327-34
42. Zagars G, Pollack A. Serum testosterone levels after external beam radiation for clinically localized prostate cancer. Int J Radiat Oncol Biol Phys 1997; 39 (1): 85-9
43. Wilson D, Harper M, Jensen H, et al. A prognostic index for the clinical management of patients with advanced prostatic cancer: a British Prostate Study Group Investigation. Prostate 1985; 7: 131-41
44. Ribeiro M, Ruff P, Falkson G. Low serum testosterone and a younger age predict for a poor outcome in metastatic prostate cancer. Am J Clin Oncol 1997; 20 (6): 605-8
45. Meikle AW, Smith JA, Stringham JD. Estradiol and testosterone metabolism and production in men with prostatic cancer. J Steroid Biochem 1989; 33 (1): 19-24
46. Henderson BE, Bernstein L, Ross RK, eta l. The early *in utero* oestrogen and testosterone environment of blacks and whites: potential effects on male offspring. Br J Cancer 1988; 57 (2): 216-8
47. Yunda I, Imshinetskaya L. Testosterone excretion in chronic prostatis. Andologia 1997; 9 (1): 89-94
48. Barabanov L. The function of the hypothesis, gonads and adrenal cortex during treatment of patients with gonorrhoea complicated prostatis. Vestn Dermatol Venerol 1989; 7: 69-74
49. Adamopouls D, Lawrence D, Vassilopoulos P. Pituitary testicular interrelationships in mumps orchitis and other viral infections. BMJ 1978; 1 (6121): 1177-80
50. Aiman J, Brenner P, MacDonaol P. Androgen and oestrogen production in elderly men with gynaecomastia and testicular atrophy after mumps orchitis. J Clin Endocrinol Metab 1980; 50 (2): 380-6
51. Anonymous. Breast cancer and hormone replacement therapy: collaborative reanalysis of data from 51 epidemiological studies of 52,705 women with breast cancer and 108,411 women without breast cancer. Collaborative Group on Hormonal Factors in Breast Cancer [published erratum appears in Lancet 1997 Nov 15; 350 (9089): 1484]. Lancet 1997; 350 (9084): 1047-59
52. Holli K, Isola J, Cuzick J. Low biologic aggressiveness in breast cancer in women using hormone replacement therapy. J Clin Oncol 1998; 16 (9): 3115-20
53. Murrell T. The potential for oxytocin (OT) to prevent breast cancer: a hypothesis. Breast Cancer Res Treat 1995; 35 (2): 225-9
54. Frayne J, Nicholson HD. Localization of oxytocin receptors in the human and macaque monkey male reproductive tracts: evidence for a physiological role of oxytocin in the male. Mol Hum Reprod 1998; 4 (6): 527-32
55. Holman CD, James IR, Segal MR, Armstrong BK. Recent trends in mortality from prostate cancer in male populations of Australia and England and Wales. Br J Cancer 1981; 44 (3): 340-8

Correspondence and offprints: Dr *Sarah Slater*, Department of Medical Oncology, Cancer Services Directorate, St Bartholomew's Hospital, 1st Floor, King George V Building, West Smithfield, London EC1A 7BE, England.
E-mail: e.m.davies@mds.qmw.ac.uk