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URINARY HORMONE EXCRETION IN BENIGN PROSTATIC HYPERPLASIA

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SUMMARY

The urinary excretion of LH, low polar oestrogens (oestrone + oestradiol-17β), dehydroepiandrosterone, androsteone, aetiocholanolone, pregnanediol and total 17-keto-and 17-ketogenic steriods has been determined in 30 men aged 60–84 years with benign prostatic hyperplasia and in 39 healthy men aged 60–79 years. The results show a significantly elevated oestrogen excretion in benign prostatic hyperplasia as compared with the control group. No other significant differences were found in the urinary hormone excretion. It is speculated that the increased oestrogen levels might stimulate the prostate growth mainly by facilitating the transport of peripheral testosterone into prostate cell.

INTRODUCTION

In 1933, Lacassagne[1] demonstrated that treatment of mice with oestrone caused a considerable growth of the dorsal prostatic lobes, accompanied by typical signs of prostatism. Zuckerman[2, 3] confirmed these results using monkeys as experimental animals and he was also able to demonstrate that the effect of oestrone could be reversed by simultaneous administration of testosterone propionate. Since then the role of oestrogens and the oestrogen–androgen balance in the etiology of benign prostatic hyperplasia (BPH) has been frequently discussed (for reviews, see 2–5).

The studies concerning the urinary excretion of oestrogens in BPH have hitherto failed to provide evidence for a distinct role of the oestrogens in this case. Marmorston and co-workers[6] as well as Kaufmann[7] found no significant differences in the urinary oestrogen excretion in BPH and in a control material. However, it should be pointed out that 38% of the control material in the former study had enlarged prostrate glands by rectal examination and that a less specific oestrogen method was used in the latter investigation[8]. Griffiths and co-workers have recently studied the plasma levels of unconjugated oestradiol-17β in BPH and normal males and found no significant difference in this respect[9]. However, there are indications that the levels of unconjugated oestradiol-17β in isolated plasma samples might not give a true picture of the oestrogen production in the male[10].

When considering androgens and related steroids, Marmorston and co-workers[11] found a significantly lower excretion of urinary androsterone in BPH but no differences in the excretion of dehydroepiandrosterone and aetiocholanolone. Kaufmann[7] found a significantly lower urinary excretion of conjugated testosterone and epitestosterone in BPH. The decreased urinary excretion of C_{19} steroids has led to speculations about an increased oestrogen/androgen ratio as the cause of BPH[6, 7, 10]. On the other hand, no significant differences in the plasma levels of unconjugated testosterone and 5α-dihydrostosterone have been found between BPH and normal healthy subjects[9, 12–14]. However, there are indications for a larger protein bound fraction of the unconjugated plasma androgens in BPH than in healthy males[14].

Studies on the plasma levels of LH, FSH and prolactin have hitherto not revealed any significant differences between BPH and healthy subjects[9].

The present communication deals with the results from determinations of the urinary excretion of LH, low polar oestrogens (oestrone + oestradiol-17β) and neutral C_{19} and C_{21} steroids in BPH and in normal healthy subjects. The results show a significantly elevated urinary oestrogen excretion in BPH as compared with the control group, while no significant differences were found in the excretion of the other hormones.

EXPERIMENTAL

Clinical material. The normal material consisted of 39 healthy men aged 60–79 years (mean age 66.7 $\pm$ 0.9 years). They had been admitted to the hospital for minor operations, predominantly for hernia and varices. There were no signs of prostatism and their prostates were found to be of normal size by rectal examination.

The BPH material consisted of 30 men aged 60–84 years (mean age 71.4 $\pm$ 1.2 years), operated for BPH by trasvesical adenoma enucleation. With the exception of one specimen of 12 g, the weight of the adenomas ranged from 30 to 160 g (mean 67.0 $\pm$ 7.7 g).

In the BPH as well as the normal material values for haemoglobin, Na^+, K^+, sedimentation rate, urinary residual nitrogen and urinary sediment were

normal. Subjects in which renal, hepatic or biliary malfunction was confirmed or suspected were not included in the study. There was no evidence of endocrinological disorder and all patients were free from medications. 24 h urine samples were collected preoperatively.

Abbreviations and trivial names. Aetiocholanolone: 3α-hydroxy-5β-androstan-17-one; Androsterone: 3α-hydroxy-5α-androstan-17-one; BPH: Benign prostatic hyperplasia; Dehydroepiandrosterone: 3β-hydroxy-5-androsten-17-one; 5α-dihydrotestosterone: 17β-hydroxy-5α-androstan-3-one; Epitestosterone: 17α-hydroxy-4-androsten-3-one; FSH: Follicle stimulating hormone; HMG: Human menopausal gonadotrophin; LH: Luteinizing hormone (synonym ICSH = Interstitial cell stimulating hormone); Oestradiol-17β: 1,3,5, (10)-oestratriene-3,17β-diol; Oestrone; 3-hydroxy-1,3,5 (10)-oestratrien-17-one; Oestrone sulphate: 3-sulphoxy-1,3,5 (10-oestratrien-17-one; Pregnanediol: 5β-pregnane-3α, 20α-diol; Testosterone: 17β-hydroxy-4-androsten-3-one.

METHODS

LH was measured by the radioimmunosorbent technique of Wide and co-workers[15] and was expressed as IU of the 2nd International Reference HMG Preparation/24 h. Low polar oestrogens were determined by the method of Carlström & Furuhjelm[16]. Fractionated 11-deoxy-17-ketosteroids (androsterone, aetiocholanolone and dehydroepiandrosterone) and pregnandiol were determined by a combination of the methods of Carlström *et al.* and Carlström and Furuhjelm[17, 18]. 17-ketogenic steroids were assayed according to Birke *et al.*[19] and total 17-ketosteroids by the method of Vestergaard[20].

RESULTS

The results from the hormone analyses are given in Table 1. The only significant difference between the two given groups was found in the excretion of the low polar oestrogens which was higher in the BPH group (mean 5.2 ± 0.5, range 1–11 μg/24h) than in the control group (mean 3.2 ± 0.3, range 1–7 μg/24h) (highly significant, $P < 0.001$). In accordance with previous findings [11] the excretion of androsterone and the 5α/5β-ratio was numerically lower in the BPH than in healthy subjects. This difference was, however, not statistically significant in the present study.

No significant correlation was found between the prostate weight and the urinary excretion of any of the hormones.

DISCUSSION

The present results give additional support to the hypothesis involving an oestrogen/androgen imbalance in favour of the oestrogens as the cause of BPH. While other authors have stressed the declined androgen levels as a cause of this imbalance[6, 7, 11], our results indicate an additional contribution of an elevated oestrogen production. It has been shown in a previous report from this laboratory[10] that the urinary excretion of low polar oestrogens in the human male remains constant at 5.2 ± 0.4 μg/24h (mean) from 20 to 59 years, but declines abruptly at approximately 60 years to 3.2 ± 0.3 μg/24h (mean) and remains rather constant at this level to at least 80 years. It is well known from several studies that the androgen production in the male declines continuously with increasing age. Thus the appearance of BPH at approximately 60 years might be related to a persisting high oestrogen level combined with a decline in the androgen production.

At present one can not exclude the possibility that the increased urinary oestrogen excretion is a phenomenon which is not directly related to the BPH, but rather a consequence of other metabolic changes more closely involved in the etiology of BPH. Hydroxylations and conjugation in the liver and the biliary and enterohepatic circulation play a vital role in the metabolism and excretion of oestrogens and an impaired liver function or decreased biliary excretion might lead to changes in the urinary excretion and in the pattern of urinary metabolites[21–25]. However, there were no indications for any hepatic or bili-

Table 1. Urinary hormone excretion in normal healthy males and in males with benign prostatic hyperplasia (BPH)

	Normal healthy males Age 66.7 ± 0.9 years (60–79); N = 39	BPH Age 71.4 ± 1.2 years (60–84); N = 30	Significant difference
LH, IU/24h	40.9 ± 3.2 (8–95)	47.2 ± 5.5 (8–144)	n.s.
Low polar oestrogens, μg/24h	3.2 ± 0.3 (1–7)	5.2 ± 0.5 (1–11)	$P < 0.001$
Total 17-ketosteroids, mg/24h	5.67 ± 0.33 (2.7–13.0)	6.19 ± 0.53 (2.1–15.2)	n.s.
11-desoxy-17-ketosteroids, mg/24h	3.07 ± 0.23 (0.5–7.5)	2.74 ± 0.23 (0.8–5.5)	n.s.
Dehydroepiandrosterone, mg/24h	0.11 ± 0.02 (0.05–0.70)	0.12 ± 0.03 (0.05–0.60)	n.s.
Androsterone, mg/24h	1.71 ± 0.14 (0.3–4.1)	1.48 ± 0.12 (0.5–2.7)	n.s.
Aetiocholanolone, mg/24h	1.22 ± 0.12 (0.1–3.4)	1.28 ± 0.14 (0.3–3.0)	n.s.
5α:5β-ratio	1.78 ± 0.17 (0.59–5.00)	1.58 ± 0.18 (0.37–4.00)	n.s.
Pregnandiol, mg/24h	0.14 ± 0.02 (0.05–0.60)	0.22 ± 0.04 (0.05–0.80)	n.s.
17-ketogenic steroids, mg/24h	7.81 ± 0.54 (3.6–16.2)	8.67 ± 0.50 (1.8–13.3)	n.s.

ary malfunction in the BPH as well as in the control material used in the present study.

An incresed oestrogen production caused by aromatization of androgens in the hyperplastic prostate itself seems less likely. As far as we know from the literature, hydroxylation of C_{19} steroids in positions 2β, 6 and 7 are the only oxygenase catalyzed steroid transformations reported to occur in prostatic tissue[26].

Several studies *in vitro* and in animals have shown that oestrogens affect the prostate gland in different ways and that a dose-effect relationship seems to exist in this respect. Thus stimulatory or synergistic effects together with androgens have been demonstrated on the entry and uptake of androgens and on the growth of the prostrate at oestrogen concentrations below 4×10^{-7} M or oestrogen/androgen ratios less than 2[27–30]. Observations concerning inhibitory or antagonistic effects seem to be restricted to higher oestrogen levels or oestrogen/androgen ratios [27, 29–36]. It should be kept in mind that the concentration of biologically active oestrogens (oestrone + oestradiol-17β + oestrone sulphate) in peripheral human male plasma is normally less than 3.5×10^{-9} M and the maximal oestrogen/androgen ratio less than 10^{-1} (calculated from the data in references 13, 37–39).

Oestrogen binding macromolecules have been demonstrated in the cytosol and the nucleus of the prostrate cell, although the existence of nuclear oestrogen receptors has been disputed[36, 40–44]. Direct interaction with these macromolecules might be one possible mode of action of oestrogens on the prostrate cell, analogous to a speculated mechanism for the testosterone stimulated growth of the uterus [45]. However, the role of the oestrogen binding macromolecules in the prostate is still not clear.

Several mechanisms might be speculated for a possible synergistic action between oestrogens and androgens on the prostrate cell. It is known that hyperplastic human prostates are far richer on the terminal androgenic steroid 5α-dihydrotestosterone than normal glands[46]. As far as we know from the literature, no differences between BPH and normal males have been found neither in the prostate cytosol 5α-reductase activity[46], nor in the peripheral plasma levels of 5α-dihydrotestosterone, testosterone and 4-androstene-3,17-dione[9, 12–14]. The higher levels of 5α-dihydrotestosterone in hyperplastic tissue might therefore be attributed to an increased entry and uptake of peripheral androgens into the cells. Giorgi and coworkers have demonstrated that oestradiol-17β in moderate concentrations ($1.1–2.2 \times 10^{-7}$ M) increases the entry and uptake of androgens in human prostatic tissue [30, 55]. This is in agreement with recent results of Lee and co-workers [47] who found that administration of oestradiol valerate to rats *in vivo* caused an accumulation of testosterone and 5α-dihydrotestosterone in the prostate and seminal vesicles. A facilitation of the transport of peripheral testosterone into the prostate cell might therefore be an important mechanism for a synergistic action of oestrogens on the androgen-stimulated prostate growth.

Studies on the effects of oestrogens on the transformation of testosterone into 5α-dihydrotestosterone by the cytosol 5α-reductase have yielded conflicting results. Stimulatory or insignificant effects have been reported for low oestrogen concentrations[35, 48–50] while higher oestrogen levels cause inhibitory effects[34, 35, 50, 51]. However, the 5α-reduction of testosterone might not be the limiting step in the formation of 5α-dihydrotestosterone[46, 52, 53].

Finally, one might speculate about a stimulatory effect of oestrogens upon the entry of the 5α-dihydrotestosterone receptor into the prostate cell nuclei, analogous to the effect of 5α-dihydrotestosterone on the accumulation of oestrogen receptor in the nuclei of uterine tissue[45, 54]. However, such a presumptive oestrogen accumulated 5α-dihydrotestosterone receptor might have empty binding sites as has been shown for the 5α-dihydrotestosterone accumulated oestrogen receptor in uterine nuclei[45], and might therefore be more or less inactive.

At present all hypotheses discussed above must be regarded as speculative. If, however, the oestrogens are directly involved in the etiology of BPH according to one of these or to some alternative mechanism, clinical studies with antioestrogens would be of great interest.

REFERENCES

1. Lacassagne A.: *C. R. Soc. biol. Paris* **113** (1933) 590–592.
2. Zuckerman S.: *Proc. Roy. Soc. Med.* **29** (1935–36) 1557–1568.
3. Zuckerman S.: *The Lancet* **231** (1936) 1259–1262.
4. Scott W. W.: *J. Urol.* **70** (1953) 477–488.
5. Grayhack J. T.: *In Urological Research Papers presented in honor of W. W. Scott.* Plenum Press, New York & London 1972, pp 39–49.
6. Marmorston J., Lombardo L. J., Myers S. M., Gierson H., Stern E. and Hopkins C. E.: *J. Urol.* **93** (1965) 287–295.
7. Kaufmann J.: *Z Urol.* **4** (1968) 229–250.
8. Ittrich G.: *Hoppe-Seylers Z. physiol. Chem.* **312** (1958) 1–14.
9. Griffiths K.: *Brit. J. Cancer* **30** (1974) 187–188.
10. Sköldefors H., Carlström K. and Furuhjelm M.: *Acta Obstet. Gynaec. scand.* **54** (1975) 89–90.
11. Marmorston J., Lombardo L. J., Myers S. M., Gierson H., Stern E. and Hopkins C. E.: *J. Urol.* **93** (1965) 276–286.
12. Becker H., Klosterhalfen H. and Voigt K. D.: *Urol. int.* **28** (1973) 350–355.
13. Mahoudeau J. A., Delasalle A. and Bricaire H.: *Acta endocr., Copenh.* **77** (1974) 401–407.
14. Horst H.-J., Becker H. and Voigt K. D.: *Steroids* **23** (1974) 833–844.
15. Wide L., Nillius S.-J., Gemzell C. and Roos P.: *Acta endocr., Copenh.* Suppl. **174** (1973).
16. Carlström K. and Furuhjelm M.: *Acta Obstet. Gynaec. scand.* **50** (1971) 259–267.

17. Carlström K., Furuhjelm M., Ingelman-Sundberg A. and Lunell N.-O.: *Int. J. Gynecol. Obstet.* **7** (1969) 124–135.
18. Carlström K. and Furuhjelm M.: *Gynecol. Prat.* **21** (1970) 443–451.
19. Birke G., Diczfalusy E. and Plantin L.-O.: *J. clin. Endocr. Metab.* **18** (1958) 736–754.
20. Vestergaard P.: *Acta endocr., Copenh.* **8** (1951) 193–214.
21. Adlercreutz H.: *Acta endocr., Copenh.* Suppl. **72** (1962).
22. Adlercreutz H. and Luukkainen T.: *Acta endocr., Copenh.* Supp. **124** (1968) 101–140.
23. Adlercreutz H.: *J. Endocr.* **46** (1970) 129–163.
24. Taylor W.: *Vit. Horm.* **29** (1971) 201–285.
25. Pentikäinen P. J., Pentikäinen L. A., Azarnoff D. L. and Dujovne C. A.: *Gastroenterology* **69** (1975) 20–27.
26. Ofner P., Vena R. L. and Morfin R. F.: *Steroids* **24** (1974) 261–279.
27. Brehmer B., Marquardt H. and Madsen P. O.: *J. Urol.* **108** (1972) 890–896.
28. Grayhack J. T.: *Endocrinology* **77** (1965) 1068–1074.
29. Karr J. P., Kirdani R. Y., Murphy G. P. and Sandberg A. A.: *Life Sci.* **15** (1974) 501–513.
30. Giorgi E. P., Stewart J. C., Grant J. K. and Shirley I. M.: *Biochem. J.* **126** (1972) 107–121.
31. Ban R. W., Cooper J. F., Imfeld H. and Foti A.: *Invest. Urol.* **11** (1974) 308–311.
32. Mahwinney M. G., Schwartz F. L., Thomas J. A. and Lloyd J. W.: *Invest. Urol.* **12** (1974) 17–22.
33. Davies P. and Griffiths K.: *J. Endocr.* **59** (1973) 367–368.
34. Farnsworth W. E.: *Invest. Urol.* **6** (1969) 423–427.
35. Jenkins J. S. and McCaffrey V. M.: *J. Endocr.* **63** (1974) 517–526.
36. Fraser H. M., Mitchell A. J. H., Anderson C. K. and Oakey R. E.: *Acta endocr., Copenh.* **76** (1974) 773–782.
37. Loriaux D. L., Ruder H. J. and Lipsett M. B.: *Steroids* **18** (1971) 463–472.
38. Rubens R., Dhont M. and Vermeulen A.: *J. clin. Endocr. Metab.* **39** (1974) 40–45.
39. Vermeulen A., Rubens R. and Verdonck L.: *J. clin. Endocr. Metab.* **34** (1972) 730–734.
40. Unehjem O.: *In Research on Steroids* (Edited by M. Finkelstein, A. Klopper, C. Conti and C. Cassano). Pergamon Press, Oxford and New York, Vol. 4. (1970) pp. 139–143.
41. Baulieau E. E., Jung I., Blondeau J. P. and Robel P.: *Adv. Biosci.* **7** (1971) 179–191.
42. McCann S., Görlich L., Janssen U. and Jungblut P. W.: In Excerpta Medica International Congress Series. (Edited by V. H. T. James) **210** (1970) 150.
43. Fang S. and Liao S.: *J. biol. Chem.* **246** (1971) 16–24.
44. Armstrong E. G. and Bashirelahi N.: *Biochem. biophys. Res. Commun.* **61** (1974) 628–634.
45. Ruh T. S., Wassilak S. G. and Ruh M. F.: *Steroids* **25** (1975) 257–273.
46. Siiteri P. K. and Wilson J. D.: *J. clin. Invest.* **49** (1970) 1737–1745.
47. Lee D. K. H., Bird C. E. and Clark A. F.: *Steroids* **26** (1975) 137–147.
48. Lee D. K. H., Bird C. E. and Clark A. F.: *J. steroid Biochem.* **5**, (1974) 609–617.
49. Danutra V., Harper M. and Griffiths K.: *J. Endocr.* **59** (1973) 539–544.
50. Altwein J. E., Rubin A., Klose K., Knapstein P. and Orestano F.: *Urologie* **A13** (1974) 41–46.
51. Tan S. Y., Antonipillai I. and Murphy B. E. P.: *J. clin. Endocr. Metab.* **39** (1974) 936–941.
52. Grant J. K. and Giorgi E. D.: *J. steroid Biochem.* **5** (1974) 949–953.
53. Bruchovsky N. and Wilson J. D.: *J. biol. Chem.* **243** (1968) 2012–2021.
54. Rochefort H., Lignon R. and Capony F.: *Biochem. biophys. Res. Commun.* **47** (1972) 662–670.
55. Giorgi E. P., Moses T. and Grant J. K.: *J. Endocr.* **61** (1974) 17.