J Int Med Res (1974) 2, 439

The Effect of Spironolactone on Plasma Levels and Excretion of Testosterone and Oestrogens in the Urine in Males

(A Preliminary Report)

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Two week's treatment with spironolactone (100 mg daily) was found to result in statistically significant decrease in plasma nonconjugated testosterone concentration associated with a significant increase in plasma LH activity in healthy males. Excretion of three main oestrogens (oestrone, oestradiol–17β and oestriol) in urine was significantly increased and the excretion of 17-ketosteroids decreased following treatment with spironolactone. Plasma levels of nonconjugated oestrogens remained unchanged as did the excretion of total testosterone in urine. The observed changes may explain some of the endocrinological side-effects of spironolactone.

Introduction

Spironolactone, a steroidal aldosterone antagonist, was initially considered to have very few endocrinological effects other than slight progestional activity (Hertz & Tullner 1958). However, it has become evident that side-effects such as gynaecomastia, decreased libido, impotence and menstrual disorders may frequently occur during spironolactone therapy (Spark & Melby 1968, Brown, Davies, Ferris & Fraser 1972, Clark 1972, Keil-Kuri & Marshall 1972, Greenblatt & Koch-Weser 1973 and Sussman 1963), suggesting a disturbed balance of the sex hormones. The mechanism of these adverse effects remains unclear although most authors have related the endocrinological effects of spironolactone to its chemical similarity with female sex hormones (Sussman 1963 and Levitt 1970).

Dymling, Nilsson & Hokfelt (1972) found a decrease in plasma testosterone (T) and excretion of 17-ketosteroids (17-KS) in urine of male subjects receiving spironolactone whereas plasma androstenedione and 17-hydroxycorticosteroid excretion in urine was not altered. These authors suggested the drug caused a decrease in testicular production of T, but had no effect on adrenal steroid production. We present data here on effects of spironolactone on plasma levels of T, three major oestrogens, luteinizing hormone (LH), and the excretion of T, oestrogens and 17-KS in the urine of healthy males.

Material and Methods

After obtaining informed consent, seven healthy males between twenty-three and thirty-four years of age were given spironolactone (Aldactone) orally, 25 mg four times

a day for fourteen days. Venous blood (8 am) and 24-hour urine collections were obtained on two consecutive days prior to and during the last two days of spironolactone dosing.

Plasma nonconjugated T was measured by the radioimmunoassay method described by Collins, Mansfield & Alladine (1972) with an antibody raised against the 3-oxime-BSA conjugate of T. Total T was determined in the urine by the same method following enzymatic hydrolysis of the glucuronide and sulfate conjugates. The coefficient of intra-assay variation was 6%. Plasma nonconjugated oestrogens were measured using the radioimmunoassay procedure detailed by Wu & Lundy (1971). Oestrone (E1), oestradiol-17β (E2), and oestriol (E3) were separated on Sephadex LH-20 columns. The oestrogens in urine were assayed by the same procedure following acid hydrolysis of the glucuronide and sulfate conjugates. Plasma LH levels were measured by radioimmunoassay (Schalch, Parlow, Boon & Reichlin 1968) in the Bio Science Laboratories (Van Nuys, Calif.) and 17-KS in urine according to Sobel *et al* (1958).Spironolactone did not interfere with any of these methods.

The means for the two days prior to spironolactone treatment were compared for difference to the post treatment period b/ the paired t-test.

Results

Treatment with spironolactone signific antly reduced the plasma T and increased th LH levels (Figure 1). The respective values were 7·29 ± 0·78 ng/ml (mean ± SE) and 8·57 ± 0·97 mIU/ml before and 6·34 ± 0·61 ng/ml ($p<0·01$) and 9·86 ± 0·86 mI J/ml ($p<0·05$) following spironolactone treat ment. No statistically significant change was ound in the plasma levels of nonconjugated ! , E2 or E3 (Table 1). Whereas the excreti n of total T in the urine did not chang the excretion of total oestrogens in urine inc ased significantly (Table 1, Figure 2). The nean oestrogen values before and following reat-ment were 7·72 ± 1·05 and 10·54 ± 1·92 μg/24 hr for E1 ($p<0·05$), 2·60 ± 0· and 3·34 ± 0·46 μg/24 hr for E2 ($p<0·0$ and 7·69 ± 0·99 and 11·75 ± 1·13 μg/24 r for E3 ($p<0·01$), respectively. The i rease was most consistent for E3, all the s jects having an elevated excretion after sp ono-lactone. The excretion of 17-KS ir urine was slightly but statistically signi antly decreased following spironolactone (T le 1). The values did not change outsi the normal range.

Table 1

Effect of spironolactone on plasma levels of oestrogens and excretion of testosterone and 17–KS in rine in healthy males

Plasma		
Hormone	*Two Days Before Treatment*	*Last Two Days Treatment*
Oestrone (pg/ml)	24·4 ± 2·5	21·2 ± 2·7
Oestradiol–17β (pg/ml)	24·1 ± 3·6	25·3 ± 3·3
Oestriol (pg/ml)	8·0 ± 0·8	8·0 ± 0·8
Urine		
Testosterone, total (μg/24 hr)	69·9 ± 17·7	68·1 ± 18·9
17–KS (mg/24 hr)	15·1 ± 1·7	13·4 ± 1·4*

Spironolactone—25 mg four times a day × 14
Mean ± SE
*$p < 0·05$

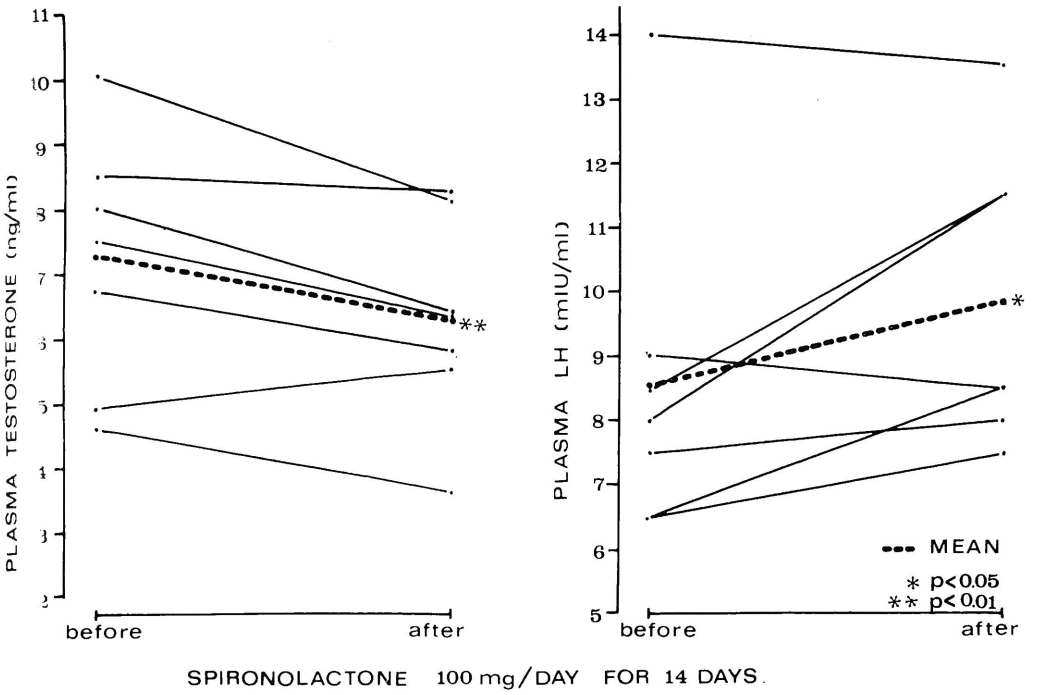

Fig *Plasma levels of nonconjugated testosterone and LH before and following treatment with spironolactone*

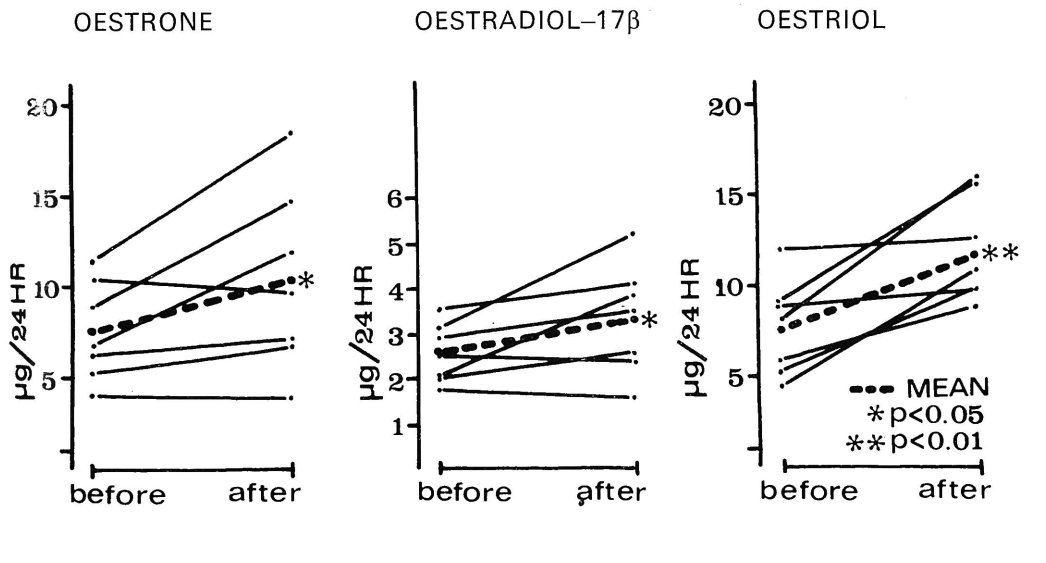

Fig 2 *Excretion of total oestrogens in urine before and following treatment with spironolactone*

Discussion

The major source of plasma T in males is the testes, less than 5% being derived from extra-testicular tissues (Horton & Tait 1966). Two other androgens, androstenedione and dehydroepiandrosterone are also secreted by the testes. These are less potent and produced in much lesser quantities than T. 5α-dihydrotestosterone, a T metabolite, is one-tenth the plasma concentration of T and due to its low affinity for the antibody used in the assay could lead to only a 2·5% overestimation of T.

Our results confirm and extend those of Dymling, Nilsson & Hokfelt (1972). Even a two-week period of treatment with therapeutic doses of spironolactone distinctly affects the balance of sex hormones in males. The mechanism by which spironolactone causes a decrease in plasma T is not clear. The change could result from a decreased rate of synthesis or increased rate of metabolism or both. A direct suppressive effect of spironolactone on testicular synthesis has been suggested by Dymling, Nilsson & Hokfelt (1972) and is supported by the findings of Stripp *et al* (1973) of a marked decrease in testicular cytochrome P-450 levels associated with decreased progesterone 17α-hydroxylase activity in the rat following treatment with 100-200 mg spironolactone/kg These doses are far in excess of the usual therapeutic doses in man. The excretion of T in urine did not change in our subjects, but may not accurately reflect testicular T production since a fraction of the T in urine is derived from the hepatic metabolism of androstenedione formed in the adrenals (Lipsett *et al* 1966).

Steroid metabolism can be accelerated by substances that induce hepatic microsomal drug oxidation. Spironolactone induces these enzymes in animals (Feller & Gerald 1971) and man (Huffman, Shoeman, Pentikäinen & Azarnoff 1973), including the 6β-hydroxylation of cortisol. Thus, spironolactone could stimulate the metabolism of T and result in a decrease in the plasma T and increase in excretion of oestrogen in urine since T is metabolized to oestrogens (Longscope, Kato & Horton 1969). Although conversion to oestrogens represents only a small part of T metabolism, an increase has been reported to occur in males with gyna eco-mastia of varying aetiologies. (Kirschn r & Taylor 1972).

Our preliminary short-term studies pr vide evidence that spironolactone has an effe t on sex hormone turnover. Studies are in pro ress to determine the responses of these horm ones to long-term chronic usage and h gher doses of spironolactone. These studies sh uld help elucidate the mechanism of the ad erse endocrinological reactions induced by s iro-nolactone.

Acknowledgement

This study was supported by United S ates Public Health Service Grant GM 1595(

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