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Effects of Continuous Levonorgestrel Treatment (Subcutaneous Capsules) on the Lipoprotein and Carbohydrate Metabolism in Fertile Women

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ABSTRACT

The lipoprotein and carbohydrate metabolism was studied during contraceptive treatment with levonorgestrel, released at a low rate (70 ug/day) from subcutaneous capsules, in five women who were treated for 180–403 days. The serum cholesterol concentration decreased slightly but significantly during treatment. This was attributable to a tendency to a reduction of cholesterol in high-density lipoproteins. The treatment did not affect the lipoprotein lipase activity in adipose tissue. The elimination rates of intravenously injected fat and glucose were unchanged, as were the serum insulin concentrations.

Thus, in this pilot study the concentration and composition of the serum lipoproteins altered to only a minor extent during treatment with levonorgestrel in a dose of 70 ug/day.

INTRODUCTION

The use of subcutaneous capsules of a silastic polymer, containing contraceptive steroids, has been investigated in contraception research (5). These devices have certain advantages over contraceptive pills. Thus the effective amount of hormone is considerably lower than the amount that has to be given in a pill. A steady plasma concentration of the hormone can be achieved. The high peaks following oral administration and the first passage through the liver can be avoided. Subjective side effects are less frequent. This report deals with the effects on the lipoprotein and carbohydrate metabolism studied in five fertile women who were treated with levonorgestrel released at a mean rate of about 70 ug/day from subcutaneous capsules. Details of the study and the effects on the ovarian steroid pattern have been described earlier (16).

MATERIALS AND METHODS

Experimental procedure

Five healthy women of normal body weight and with a mean age of 27 years (range 22-32) volunteered for the study. Sterilized (by irradiation) silastic capsules containing levonorgestrel were inserted in the subcutaneous tissue in the gluteal region one week after the start of menstruation. The capsules were removed after varying intervals and the amount of levonorgestrel remaining in the capsules was determined. The release of levonorgestrel was calculated to have varied from 39 to 100 ug/day.

The carbohydrate and lipoprotein metabolism was studied one week before the insertion of the capsules (i.e. at the start of menstruation), one week after this insertion and then after one, two and four months of treatment. The tests were repeated two to eight weeks after removal of the capsules.

Methods

Very-low-density lipoproteins (VLDL) in serum were isolated in the top fraction after preparative ultracentrifugation at a density of 1.006 in a Beckman L2-65B ultracentrifuge, using a 40.3 rotor (6). Low-density lipoproteins (LDL) were precipitated from the bottom fraction by a heparin and manganese-chloride solution (2) after the ultracentrifugation step and high-density lipoproteins (HDL) were isolated in the supernatant after precipitation. The concentrations of triglycerides and cholesterol were determined in whole serum, in VLDL and HDL and in the bottom fraction. The lipid concentrations in LDL were calculated as the differences between the concentrations in the bottom fraction and those in HDL. A Technicon Auto-Analyzer II was used for the determination of triglycerides and cholesterol (14). The accuracy and precision of the lipid determinations were checked by the Laboratory for Disease Control, Atlanta, Georgia, USA.

The concentrations of apolipoproteins (apo) B and A-I were determined in frozen serum samples on a later occasion. Rocket immuno-electrophoresis according to the method of Laurell (11), modified essentially as described by Curry (4), was used. The details of the procedure have been presented elsewhere (17).

Specimens of abdominal adipose tissue were taken by means of a needle-biopsy technique (7). One portion of adipose tissue was used for determining cell size (15) and one for determining lipoprotein-lipase activity (12). An intravenous (i.v.) fat tolerance test was performed by the method of Carlson and Rössner (3), and an i.v. glucose tolerance test as described by Ikkos and Luft (8). Serum insulin concentrations were determined by a radioimmunoassay technique (Phadebas, Pharmacia, Uppsala, Sweden).

Statistics

For each variable the hypothesis of equal means for the different time points was tested in an analysis of variance model including the main factors patients and time points. If the overall test for time points was significant, some contrasts in the time point means were tested. The level of significance was adjusted to protect against the mass-significance problem.

Results

There was a small but statistically significant reduction of serum cholesterol during treatment (Table I). This was not attributable to any change in LDL cholesterol. HDL cholesterol, on the other hand, was lower during treatment in all five subjects, but the decrease varied markedly from one individual to another. Such an interindividual variation has less impact on the calculations after a logarithmic transformation of the values. Calculated on such transformed values, the results indicated that the treatment was associated with a significant decrease of the serum HDL-cholesterol concentration. On the average HDL-cholesterol was reduced by less than 10% in four of five subjects.

TABLE I. The concentrations of triglycerides (TG) and cholesterol (Chol) (mmol/l) in very-low-density lipoproteins (VLDL), low-density lipoproteins (LDL), high-density lipoproteins (HDL) and whole serum. Mean (±SD) before (pre) and after one week (w), 1, 2 and 4 months (m) and 2 - 8 weeks after treatment stopped (post). Analysis of variance disclosed a significantly different value during treatment only for serum cholesterol ($p < 0.05$).

		VLDL		LDL		HDL		Serum	
Treatment	n	TG	Chol	TG	Chol	TG	Chol	TG	Chol
Pre	5	0.42±0.19	0.11±0.06	0.27±0.05	2.66±0.74	0.20±0.03	1.67±0.57	0.88±0.21	4.39±0.65
1 w	4	0.41±0.25	0.14±0.08	0.28±0.04	2.64±0.18	0.18±0.03	1.37±0.21	0.95±0.29	4.32±0.34
1 m	3	0.26±0.06	0.05±0.04	0.29±0.13	2.58±0.40	0.21±0.04	1.33±0.21	0.76±0.16	3.95±0.65
2 m	4	0.38±0.10	0.11±0.04	0.27±0.04	2.79±0.26	0.18±0.04	1.24±0.15	0.77±0.07	4.17±0.25
4 m	3	0.62±0.21	0.25±0.19	0.32±0.06	2.89±0.29	0.20±0.02	1.19±0.31	1.20±0.26	4.49±0.10
Post	5	0.39±0.14	0.11±0.03	0.31±0.08	3.01±0.21	0.19±0.03	1.57±0.20	0.90±0.18	4.76±0.26

TABLE II. Body weight (kg), the concentration of apolipoprotein (apo) B and A-I (arbitrary units), fasting-serum insulin (mU/l), the adipose-tissue (AT) lipoprotein-lipase activity (LPLA), mU/l, (1 mU signifies the release of one nmol of fatty acid per min), the adipocyte weight (µg), the fractional-removal rate (K-value) of the i.v. glucose-tolerance test (IVGTT) and the K_2-value (%/min) of the i.v. fat-tolerance test (IVFTT) are presented. Mean (±SD) before (pre) and after one week (w), 1, 2 and 4 months (m) and 2 - 8 weeks after treatment stopped (post). Treatment was not associated with significant changes in any variable as indicated by analysis of variance.

Treatment	n	Body weight	Apo B	Apo A-I	Insulin	AT-LPLA	K_{IVGTT}	K_{2IVFTT}
Pre	5	67.1±11.5			7.8±2.1	73.9±31.6	2.1±0.7	7.7±2.6
1 w	4	69.8±11.1			9.9±2.1	96.9±89.3	2.6±0.4	9.0±3.6
1 m	3	65.3±3.4	48±17	90±24	6.8±2.6	181.2±75.9	2.7±1.9	9.0±2.3
2 m	4	68.8±12.8	50±6	97±17	9.7±5.2	75.2±34.4	2.0±0.7	8.1±3.3
4 m	3	72.0±8.6	52±24	89±25	10.7±3.8	85.0±32.6	1.9±0.6	6.3±3.1
Post	5	67.4±10.6	52±17	94±23	11.9±3.9	112.8±59.0	1.9±0.8	6.5±2.9

The VLDL lipids and whole-serum triglycerides were not significantly affected by the treatment (Table I). Apo A-I and B did not change significantly after termination of treatment as compared with the values during treatment. Neither the adipose tissue lipoprotein lipase activity, fractional removal rates at the i.v. fat tolerance and i.v. glucose tolerance tests, nor the fasting serum insulin concentrations were altered during the treatment (Table II).

DISCUSSION

In a small pilot study like this one the possibility cannot be ruled out that some small effects, especially on parameters with a large day-to-day variability, may fail to be detected. The present results indicate that release of about 70 ug of levonorgestrel daily is associated with only minor effects on the lipoprotein metabolism. However, a small effect on the serum cholesterol was discernible, probably reflecting minor changes in the HDL-cholesterol concentration. It is well established that gestagens derived from androgens decrease HDL cholesterol in a dose-related manner (1).The release of levonorgestrel from the capsules in this study was about one-fourth of the mean release in a similar study (19) in which HDL cholesterol decreased by an average of 24% (19).

Levonorgestrel has been shown to influence the activity of hepatic lipase (9), a phospholipase involved in the catabolism of HDL. A high activity of hepatic lipase is associated with low phospholipid and cholesterol contents in HDL and a low cholesterol/protein ratio in the HDL fraction (10). In a recent study treatment with a contraceptive pill containing 150 ug of ethinyl-estradiol caused no change in HDL- and LDL-cholesterol concentrations (18). This is consistent with the finding that the estrogen component reduces hepatic lipase (9), balancing the stimulatory effect of levonorgestrel on this enzyme. However, apo A-I and B were elevated in that study by about 15% (18). This may have been an effect of ethinyl-estradiol, which has been shown to influence the synthesis of apo B in the liver (13).

In the present series the treatment inhibited ovulation but did not suppress the follicular activity, as judged by the estradiol pattern in plasma. During the treatment bursts of estrogen occurred, which increased the plasma levels of estradiol to 200-300 pg/ml (with peaks up to 750 pg/ml) for periods of up to 60 days (16). These relatively longlasting, moderately elevated levels of estradiol did not, however, influence the apolipoprotein concentrations to a significant degree.

In summary, this pilot study in five women showed a lower serum cholesterol and a tendency to a lower HDL-cholesterol level during treatment with con-

tinuously released levonorgestrel. The effects were much less pronounced with this low-dose treatment than with other contraceptive regimens with levonorgestrel, both with regard to the level of lipoprotein lipids and concerning the lipid/protein composition.

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