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The Mechanism of Action of a New Low-dosed Combined Oral Contraceptive*

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Die Wirkungsweise eines niedrig-dosierten Kombinationspräparats

Zusammenfassung. Bei zwei Gruppen von Frauen mit normalem Zyklus wurde die Wirkung des neuen niedrig-dosierten Kombinationspräparats Ovoresta M (37,5 µg Äthinyl Östradiol und 0,75 mg Lynöstrenol) auf die Gonadotropin-Freisetzung und die Follikel-Aktivität untersucht. Unter der Einnahme dieses Präparats vom 1. Zyklustag an war die Gonadotropin-Sekretion gestört und die präovulatorischen LH- und FSH-Gipfel waren unterdrückt. Im Durchschnitt waren die LH- und FSH-Spiegel etwas niedriger als im Normalzyklus; der Unterschied war jedoch nicht signifikant. In einem Fall stieg die Östradiol-Sekretion in Zyklusmitte wie in einem normalen Zyklus an, was auf eine Follikel-Aktivität schließen ließ. Obwohl ein Anstieg des Serum-Östradiols auf normale Werte beobachtet wurde, wenn mit der Einnahme des Präparats erst am Tag 10 des Zyklus begonnen wurde (bei 4/5), waren sowohl LH- und FSH-Gipfel als auch die Ovulation unterdrückt (bei 3/4). Die Hypophysen-Reaktion auf 100 µg LH-RH am Tag 21 war beeinträchtigt, wobei die LH-Freisetzung signifikant mit der Serumkonzentration des Östradiols korrelierte.

Zusammenfassend läßt sich annehmen, daß niedrig-dosierte orale Kontrazeptiva durch Störung der Follikel-Reifung und Hemmung des präovulatorischen LH-Gipfels wirken.

Schlüsselwörter: Niedrig dosierte Kombinationspräparate – Gonadotropine – Follikuläre Aktivität – LH-RH-Test

Summary. The effect of a new low-dosed combined oral contraceptive (OC) containing 37.5 µg ethinyl estradiol and 0.75 mg lynestrenol (Ovoresta M) upon gonadotropin release and follicular activity was studied in two groups of normally cyclic women. When the administration of the OC was started on day 1 of the cy-

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cle, the normal pattern of gonadotropin secretion was disrupted, and the midcycle LH and FSH peak was abolished. The mean level of LH and FSH was somewhat lower than in normal cycles, but the difference was not significant. In one case, serum estradiol rose to the level of the normal cycle indicating follicular activity. Even though there was a rise in serum estradiol to normal values when the OC was started on day 10 of the cycle (in 4/5), both the midcycle LH and FSH surge and ovulation were suppressed (in 3/4). The pituitary response to 100 μg LH-RH on day 21 was impaired. The LH-response correlated significantly with the serum estradiol concentration.

In summary, the low-dosed OC exerts its effect by interfering with follicular ripening and inhibiting the preovulatory LH surge.

Key words: Low-dosed combined OC – Gonadotropins – Follicular activity – LH-RH-test

Oral contraceptives (OC) are known to affect the secretion of gonadotropins from the pituitary, and to diminish the responsiveness of the gonadotrophs towards LH-RH in a dose-dependent manner (Aktories et al., 1976; Dericks-Tan et al., 1976). Although progestogens can interfere with the cyclic pattern of gonadotropin release, the degree of pituitary suppression by OC is clearly related to the dose of the estrogenic component in that preparations containing less than 50 μg of estrogen have less of an inhibitory effect than those with a higher dose (Dericks-Tan et al., 1976; Scott et al., 1978). This raised the question whether or not low-dosed OC would exert their effect only by inhibiting the preovulatory peak of LH and FSH, or if they would affect the basal secretion of both gonadotropins, too. In view of this we studied the effect of a new low-dosed combined OC containing 37.5 μg ethinyl-estradiol (EE_2) and 0.75 mg lynestrenol (Ovoresta M), Organon, München) upon the pituitary-ovarian axis and the pituitary response to LH-RH. In a first set of experiments, the compound was administered in the usual manner beginning on day 1 of the cycle. As the negative feedback effect of estrogens has been shown to be much less pronounced in the late follicular phase as compared to the early one (Monroe et al., 1972; Yen et al., 1972; Cargille et al., 1973), and synthetic progestogens have been shown to inhibit the production of progesterone by the corpus luteum when administered during the luteal phase (Johansson, 1971), we also investigated whether this preparation would interfere with the midcyclic gonadotropin surge and corpus luteum function, respectively, when the beginning of treatment with the OC was delayed to day 10 of the cycle.

Volunteers and Methods

Twenty-three women, aged 21 to 34 years, volunteered for the study. Written informed consent was obtained. The mean length of the cycle ranged from 24 and 32 days. The use of OC had been discontinued for at least 3 months prior to the begin of the study. A biphasic basal body temperature curve, and serum progesterone (Prog) values exceeding 3 ng/ml in the luteal phase of the control cycle were taken as evidence for ovulatory cycles.

Six women took Ovoresta M beginning on day 1 of the cycle (Group I). Five women took the preparation beginning on day 10 of the cycle (Group II). They were informed that no contraceptive effect

was to be expected at this treatment scheme. The remaining 12 women served as a control group. On day 21 of the cycle, an LH-RH stimulation-test with 100 μg LH-RH (Hoechst A.G.) was carried out in all volunteers.

Blood samples were obtained immediately before the injection of LH-RH, and 20, 40, and 60 min thereafter. Moreover, blood samples were also taken daily during the OC application and every other day before or after the OC application.

The hormone determinations were carried out by radioimmunoassay. Ovarian activity was monitored by the measurement of estradiol (E_2) and of Prog in serum. The peptide hormone standards used were immunologically calibrated against an international standard and had the following activity:

LH 1 ng 2.8 ± 0.4 mIU MRC 68/40 Mill Hill,

FSH 1 ng 2.6 ± 0.4 mIU MRC 69/104 Mill Hill.

Results

OC Application Starting on Day 1 of the Cycle

The effect of the administration of the low-dosed combined OC upon gonadotropin secretion and ovarian function is shown in Fig. 1. The frames on the right of the diagram depict the results of the LH-RH stimulation test carried out on day 21. The midcyclic LH and FSH peak was clearly suppressed in all women. Contrary to that, the basal pattern of LH and FSH secretion was not significantly affected in four of the six volunteers in that the values tended to be low but to be still within the range of normally cyclic women (Follicular phase: LH = 0.8–3.5 ng/ml, FSH = 0.9–4.3 ng/ml; Luteal phase: LH = 1.0–4.2 ng/ml, FSH = 0.8–3.2 ng/ml). The remaining two women showed a normal basal gonadotropin secretion. A "rebound-like" phenomenon was discernible after cessation of OC application in five of six women.

In one woman (J. H.), the secretion of E_2 resulted in even higher serum levels at midcycle than that normally observed in cyclic women. Otherwise, there was not much evidence of ovarian activity, as E_2 rose only transiently to values in excess of 100 pg/ml in two subjects. Corpus luteum formation did not occur as serum Prog remained at values below 1 ng/ml throughout the period of treatment.

OC Application Starting on Day 10 of the Cycle

When the application of the OC was not begun before day 10 of the cycle, an effect upon gonadotropin secretion similar to that seen in the subjects taking the OC in the conventional manner was observed at the corresponding time of the cycle (Fig. 2). In four of five volunteers comprising Group II, no midcyclic rise in serum LH and FSH was demonstrable. A somewhat blunted LH peak (6.1 ng/ml) coinciding with a normal FSH peak (4.2 ng/ml) was observed in the fifth woman (J. K.). As had been shown in Group I, the serum level of LH and FSH tended to be low but still remained within the limits of the normal range. Contrary to Group I, ovarian function proved to be much less inhibited. Even though a rise in serum E_2 mimicking that of the normal ovulatory cycle was seen in all but one of the subjects, the maximal values actually exceeding the upper limit of the norm in two cases, ovulation occurred only once. In this women (J. K.), a small LH surge on day 10 of the cycle was followed by two Prog peaks of equal height which coincided with elevations of serum E_2, the second one reaching more than

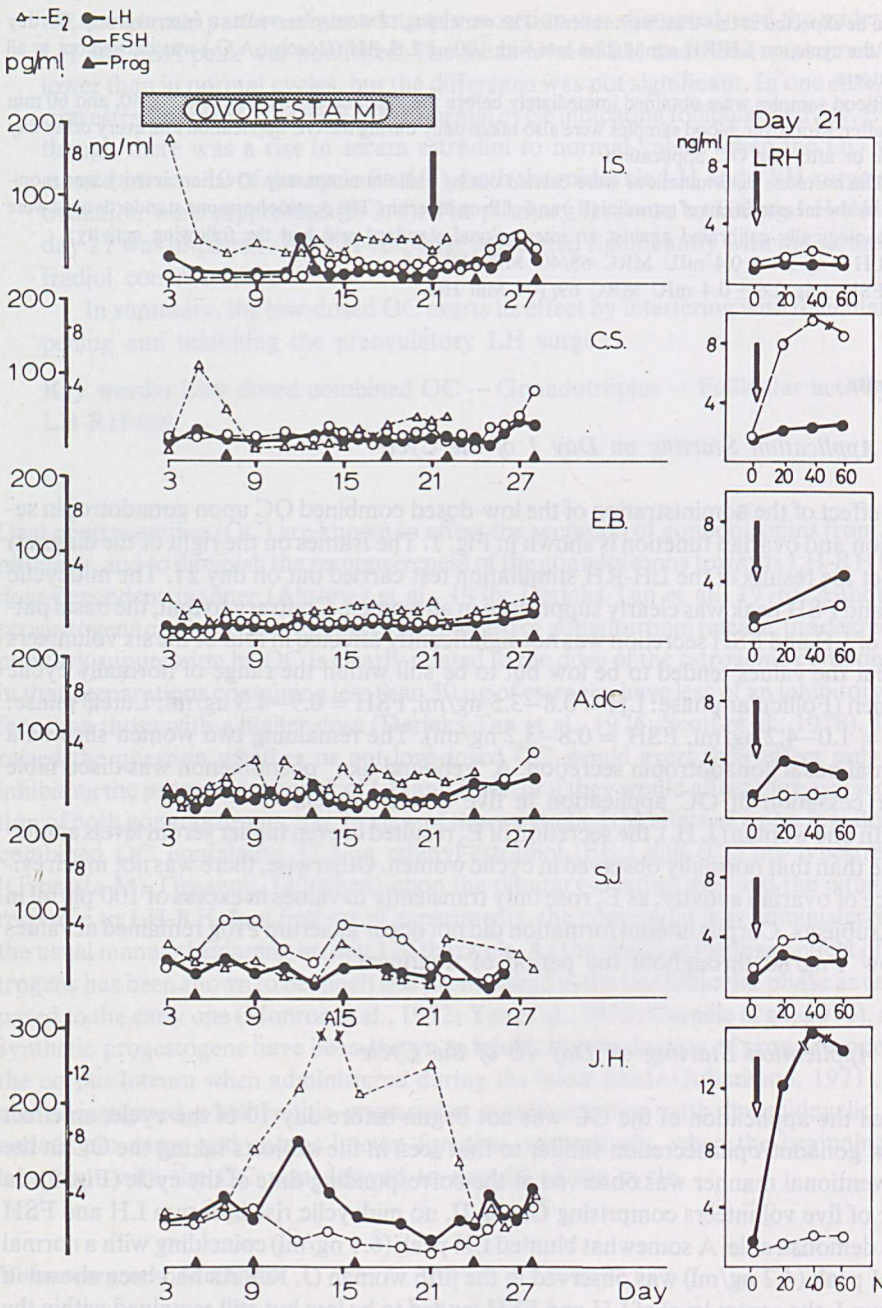

Fig. 1. The effect of daily treatment with the OC containing 37.5 μg ethinyl estradiol plus 0.75 mg lynestrenol in normally cyclic women starting on day 1 of the cycle upon the secretion of LH, FSH, E_2, and Prog. The LH-RH stimulation test (100 μg i.v.) on day 21 is shown in the right frame

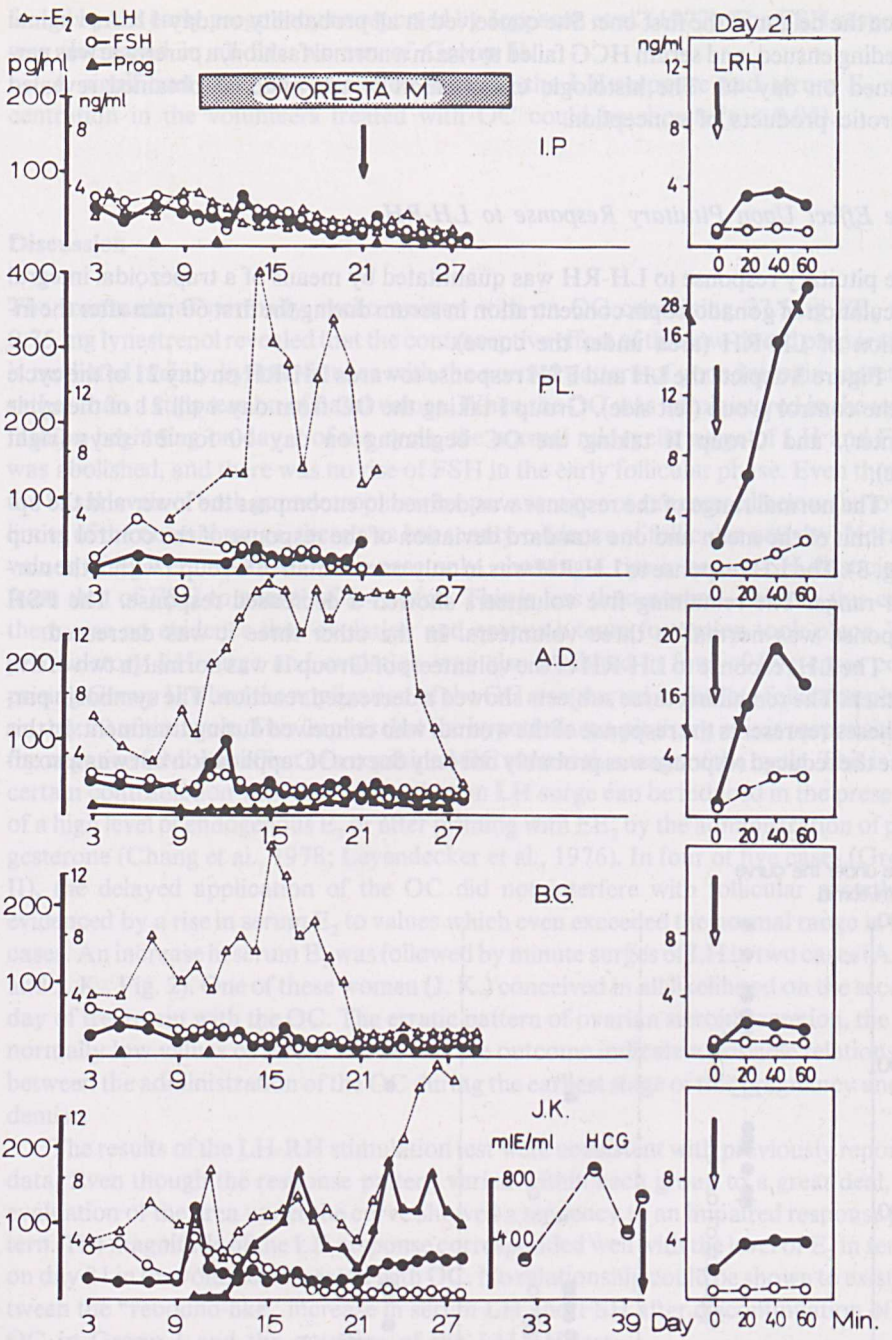

Fig. 2. The effect of the OC treatment starting on day 10 of the cycle upon serum LH. FSH, E_2, and Prog. The LH-RH stimulation test on day 21 is shown in the right frame

twice the height of the first one. She conceived in all probability on day 11. As vaginal bleeding ensued, and serum HCG failed to rise in a normal fashion, a curettage was performed on day 40. The histologic examination of the material obtained revealed necrotic products of conception.

The Effect Upon Pituitary Response to LH-RH

The pituitary response to LH-RH was quantitated by means of a trapezoidal integral calculation of gonadotropin concentration in serum during the first 60 min after the injection of LH-RH (area under the curve).

Figure 3 depicts the LH and FSH response towards LH-RH on day 21 of the cycle of the control group (left side), Group I taking the OC from day 1 till 21 of the cycle (center), and Group II taking the OC beginning on day 10 for 21 days (right side).

The normal range of the response was defined to encompass the lower and the upper limit of the mean and one standard deviation of the response of the control group (Fig. 3). The LH response to LH-RH was in only one woman of Group I within the normal range. The remaining five volunteers showed a decreased response. The FSH response was normal in three volunteers. In the other three it was decreased.

The LH response to LH-RH of the volunteers of Group II was normal in two of five women. The remaining three subjects showed a decreased reaction. The symbol in parentheses represents the response of the woman who conceived during treatment. In this case the reduced response was probably not only due to OC application but was also af-

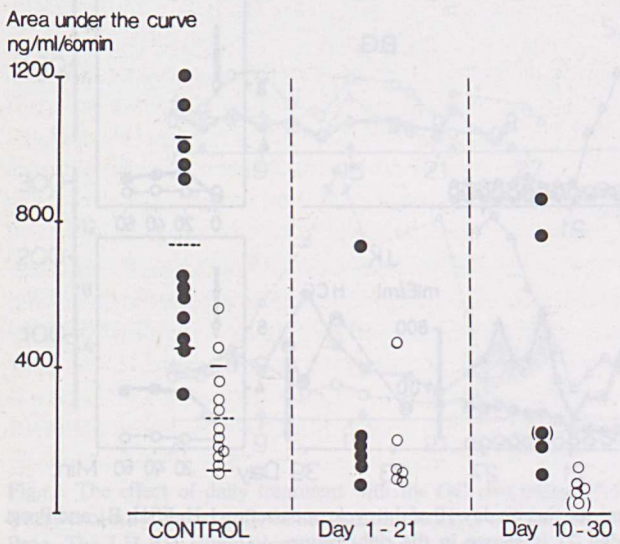

Fig. 3. The LH (●) and FSH (○) response to 100 μg LH-RH carried out on day 21 of the cycle in the control group (left), Group I taking the OC starting on day 1 (center), and Group II starting on day 10. The response was expressed as the gonadotropin released within 60 min after the LH-RH injection. The horizontal lines in the control group represent the mean ± SD

fected by the early pregnancy as reported by Jeppsson et al. (1977). The FSH response was decreased in all five women of Group II.

A significant positive correlation between the LH-response and serum E_2 concentration in the volunteers treated with OC could be shown ($p < 0.05$).

Discussion

The treatment of normally cyclic women with an OC containing 37.5 μg EE_2 and 0.75 mg lynestrenol revealed that the contraceptive effect of this low-dosed preparation is mediated mainly by interference with the normal pattern of gonadotropin secretion rather than a suppression of basal values. When the OC was administered in the usual fashion beginning on day 1 of the cycle, the normal midcyclic surge of LH and FSH was abolished, and there was no rise of FSH in the early follicular phase. Even though the basal levels of both gonadotropins were per average not suppressed below the lower limits of the normal range, there was but scanty evidence of follicular activity. Normal values of E_2 were attained in one case only, showing a rise in serum LH dissociated from that of FSH to a level of 8.4 ng/ml. This is less than normal. Even in this case, there was no evidence that ovulation and corpus luteum formation took place. The preovulatory LH-surge and ovulation was also inhibited in four of five cases comprising Group II when the application of the OC was started in the late follicular phase on day 10 of the cycle. This implies that the hypothalamo-pituitary axis is responsive to the negative feedback effect of a combined OC even at this stage of the cycle. This is in a certain contradiction with the finding that an LH surge can be induced in the presence of a high level of endogenous E_2 or after priming with EE_2 by the administration of progesterone (Chang et al., 1978; Leyendecker et al., 1976). In four of five cases (Group II), the delayed application of the OC did not interfere with follicular growth as evidenced by a rise in serum E_2 to values which even exceeded the normal range in two cases. An increase in serum E_2 was followed by minute surges of LH in two cases (A. D. and J. K., Fig. 2). One of these women (J. K.) conceived in all likelihood on the second day of treatment with the OC. The erratic pattern of ovarian steroid secretion, the abnormally low values of serum HCG, and the outcome indicate a possible relationship between the administration of the OC during the earliest stage of this pregnancy and its demise.

The results of the LH-RH stimulation test were consistent with previously reported data. Even though the response pattern varied within each group to a great deal, the evaluation of the area under the curve showed a tendency to an impaired response pattern. The magnitude of the LH response corresponded well with the level of E_2 in serum on day 21 in the volunteers treated with OC. No relationship could be shown to exist between the "rebound-like" increase in serum LH and FSH after discontinuation of the OC in Group I and the outcome of the LH-RH-test.

We are concluding that the high degree of contraceptive effectiveness of this low-dosed OC can mainly be ascribed to the following facets of its gonadotropin-inhibiting activity: It interferes with follicular ripening and inhibits the preovulatory surge of LH. The preparation could even be shown to be capable of inhibiting ovulation quite effectively (four of five cases) under rather adverse conditions, i.e., when its

application was started but a few days prior to the expected date of ovulation, and in the presence of a ripening follicle.

When the OC was, however, taken in the usual fashion beginning on day 1 of the cycle, there were subtle changes in the secretory pattern of LH and FSH which were clearly reflected in a marked impidement of follicular activity in most cases.

References

Aktories, K., Krög, W., Dericks-Tan, J. S. E., Jürgensen, O., Taubert, H.-D.: Die Beeinflussung des Ovarialzyklus durch verschiedene Typen hormonaler Kontrazeptiva. Geburtsh. Frauenheilk. **36,** 318–326 (1976)

Cargille, C. M., Vaitukaitis, J. L., Bermudez, J. A., Ross, G. T.: A differential effect of ethinyl estradiol upon plasma FSH and LH relating to time of administration in the menstrual cycle. J. Clin. Endocrinol. Metab. **36,** 87–94 (1973)

Chang, R. J., Jaffe, R. B.: Progesterone effects on gonadotropin release in women pretreated with estradiol. J. Clin. Endocrinol. Metab. **47,** 119–125 (1978)

Dericks-Tan, J. S. E., Krög, W., Aktories, K., Taubert, H.-D.: Dose-dependent inhibition by oral contraceptives of the pituitary to relase LH and FSH in response to stimulation with LH-RH. Contraception **14,** 171–181 (1976)

Jeppsson, S., Rannevik, G., Liedholm, P., Thorell, J. I.: Basal and LRH-stimulated secretion of FSH during early pregnancy. Am. J. Obstet. Gynecol. **127,** 32–36 (1977)

Johansson, E. D. B.: Depression of the progesterone levels in women treated with synthetic gestagens after ovulation. Acta Endocrinol. (Kbh.) **68,** 779–792 (1971)

Leyendecker, G., Wildt, L., Gips, H., Nocke, W., Plotz, E. J.: Experimental studies on the positive feedback effect of progesterone, 17α-hydroxyprogesterone, and 20α-dihydroprogesterone on the pituitary release of LH. Arch. Gynäk. **221,** 29–45 (1976)

Monroe, S. E., Jaffe, R. B., Midgley Jr., A. R.: Regulation of human gonadotropins. XII. Increase in serum gonadotropins in response to estradiol. J. Clin. Endocrinol. Metab. **34,** 342–347 (1972)

Scott, J. Z., Kletzky, O. A., Brenner, P. F., Mishell, D. R.: Comparison of the effects of contraceptive steroid formulations containing two doses of estrogen on pituitary function. Fertil. Steril. **30,** 141–145 (1978)

Yen, S. S. C., Tsai, C. C.: Acute gonadotropin release induced by exogenous estradiol during the mid-follicular phase of the menstrual cycle. J. Clin. Endocrinol. Metab. **34,** 298–305 (1972)

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