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REVIEW

Ovarian function during hormonal contraception assessed by endocrine and sonographic markers: a systematic review

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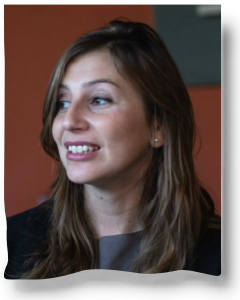

Dr Stella D'Arpe graduated from the Medical School at the University of Rome 'Sapienza' in 2007. In 2014 she completed her gynaecological and obstetrical residency training at Rome University Hospital. Currently, she is completing her PhD thesis on the clinical, endocrine and sexual impact of hormonal contraceptives comparing different dose of steroids and administration modes at Rome University Hospital.

Abstract This systematic review focuses on the literature evidence for residual ovarian function during treatment with hormonal contraceptives. We reviewed all papers which assessed residual ovarian activity during hormonal contraceptive use, using endocrine markers such as serum anti-Müllerian hormone (AMH) concentrations, FSH, LH, oestradiol, progesterone and sonographic markers such as antral follicle count (AFC), ovarian volume and vascular indices. We considered every type (oestroprogestin or only progestin) and dosage of hormonal contraceptive and every mode of administration (oral, vaginal ring, implant, transdermal patch). We performed an electronic database search for papers published from 1 January 1990 until 30 November 2015 using PubMed and MEDLINE. We pre-selected 113 studies and judged 48 studies suitable for the review. Most studies showed that follicular development continues during treatment with hormonal contraceptives, and that during treatment there is a reduction in serum concentrations of FSH, LH and oestradiol, and also a reduction in endometrial thickness, ovarian volume and the number and size of antral follicles. The ovarian reserve parameters, namely AFC and ovarian volume, are lower among users than among non-users of hormonal contraception; regarding the effect of hormonal contraception on AMH, there are still controversies in the literature.

KEYWORDS: follicular development, hormonal contraception, hormone-free interval, ovarian activity, ovarian function

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Introduction

The most important effect of hormonal contraception is the inhibition of hypothalamo-pituitary axis causing a decrease in FSH and LH, leading to the suppression of follicular activity and ovulation. In the last 50 years, the composition of hormonal contraceptives has undergone several modifications in order to reduce as much as possible the side effects, and to increase the compliance of women, while preserving contraceptive efficacy. Scientific literature shows that reducing the dose of oestrogen of combined contraceptives, to minimize its adverse effects, is associated with a decrease in pituitary gonadotrophin secretion, particularly during the hormone-free interval (HFI) or following missed doses, resulting in greater follicular development (Baerwald and Pierson, 2004). In this regard, several studies have evaluated the residual ovarian function during treatment with hormonal contraceptives, comparing different doses of steroids, duration of the HFI, and administration schemes. Many authors tried to study the ovarian function using endocrine markers like serum anti-Müllerian hormone (AMH) concentrations, FSH, LH, oestradiol, progesterone and sonographic markers such as antral follicle count (AFC), ovarian volume and vascular indices. The aim of this review is to give a complete evaluation of residual ovarian activity during hormonal contraceptive use.

Materials and methods

The present systematic review included all scientific articles which assessed residual ovarian activity during hormonal contraceptive use. All articles reporting ovarian activity as evaluated with either sonographic parameters or biochemical parameters were included. Studies were excluded if reporting only biochemical parameters. Every type and dosage (oestroprogestin [EP] or only progestin) of hormonal contraceptive and every mode of administration (oral, vaginal ring, implant, transdermal patch) were considered.

Included studies were randomized clinical trials, prospective controlled studies, prospective cohort studies or retrospective studies with a sample ≥30 patients in good health. Studies about women in breastfeeding or overweight were excluded. Only articles written in English were included.

An electronic database search was performed using PubMed and MEDLINE for the identification of articles published from 1 January 1990 to 30 November 2015, using the combination of the following search terms: contraceptives, hormonal contraceptives, contraception, hormonal contraception, steroid contraception, oral contraception, oestroprogestins, follicle, follicle development, follicular development, hormone-free interval, ovarian activity, ovarian function, AMH, LH, FSH, follicle cysts, ovarian cysts, ovulation, ultrasound. Three investigators independently conducted this search. After the search, all relevant studies were retrieved based on the title and the abstract content, and their reference lists were checked manually to identify additional potential studies. The full text of the identified papers was analysed independently by three investigators with the purpose of determining whether or not to include the article in the systematic review. In cases of incomplete data, studies were excluded. In cases of disagreement in the review process, consensus was achieved through the involvement of other investigators.

One hundred and thirteen studies were pre-selected after the electronic search based on the article title and abstract, and after a manual search of the reference lists of the full articles. After reading of the full text, a total of 67 articles were excluded. A total of 46 studies were therefore judged suitable for the review (**Figure 1**).

Ovarian function during combined contraceptive use

It has been shown that oestrogens and progestins at the concentrations above physiological level, produce a negative feedback effect on the hypothalamo-pituitary axis (Wan et al., 1981). Presumably reduced gonadotrophin-releasing hormone (GnRH), FSH and LH concentrations inhibit ovarian follicular growth and consequently suppress ovulation and conception. The progestins have been shown to prevent the LH surge and ovulation (Barnhart et al., 1997; Tafurt et al., 1980). The oestrogens are believed to suppress the development of preantral and medium-sized antral follicles in primates (Koering et al., 1991, 1994), presumably through suppression of FSH secretion. Moreover, oestrogens have the function of improving satisfaction of patients avoiding irregular bleeding patterns. However, during the use of EP, residual follicular activity has been shown to persist. In women using a combined oral contraceptive (COC), the degree of follicular activity seems to depend on the dose of ethinyl oestradiol (EE) rather than on the dose and type of progestin. (Fauser and Van Heusden, 1997; Spellacy et al., 1980). In fact, during low EE dose formulations use, greater numbers and bigger diameter of follicles are observed (Teichmann et al., 1995). Therefore, the decrease of oestrogen dose in COC could reduce the degree of hypothalamo-pituitary ovarian suppression especially after missed doses, or during HFI. Indeed, in women using COC, follicular growth appears to take place more frequently during HFI, where there is a loss of endocrine suppression (Rabe et al., 1997).

Three prospective studies examined ovarian function in women using a single formulation of COC (Deb et al., 2012; Hoogland and Skouby, 1993; Spona et al., 2010). A prospective cohort study compared ovarian reserve markers between users and non-users of hormonal contraception (Bentzen et al., 2012).

In a prospective study Deb et al. (2012) analysed sonographic and endocrine markers in 34 women who had been using a COC containing 30 μg EE + 150 μg levonorgestrel (LNG) with HFI for a period longer than one year, compared with 36 controls who had not used a COC within the previous year. The COC group had a significantly lower number of antral follicles measuring ≥6 mm ($P < 0.001$) and significantly lower ovarian volume, ($P < 0.001$); the vascular indices were also lower in the COC group than in controls but the number of small antral follicles measuring 2–6 mm was similar among the two groups. As regards endocrine markers, FSH, LH and oestradiol concentrations were significantly lower in the COC group ($P < 0.05$), but serum AMH concentrations were not statistically different between the two groups.

In another prospective study the authors evaluated oestradiol values and performed ultrasound scans in 87 women that used 30 μg EE + 75 μg gestodene (GSD) for two cycles. They monitored the ovarian activity by describing follicle-

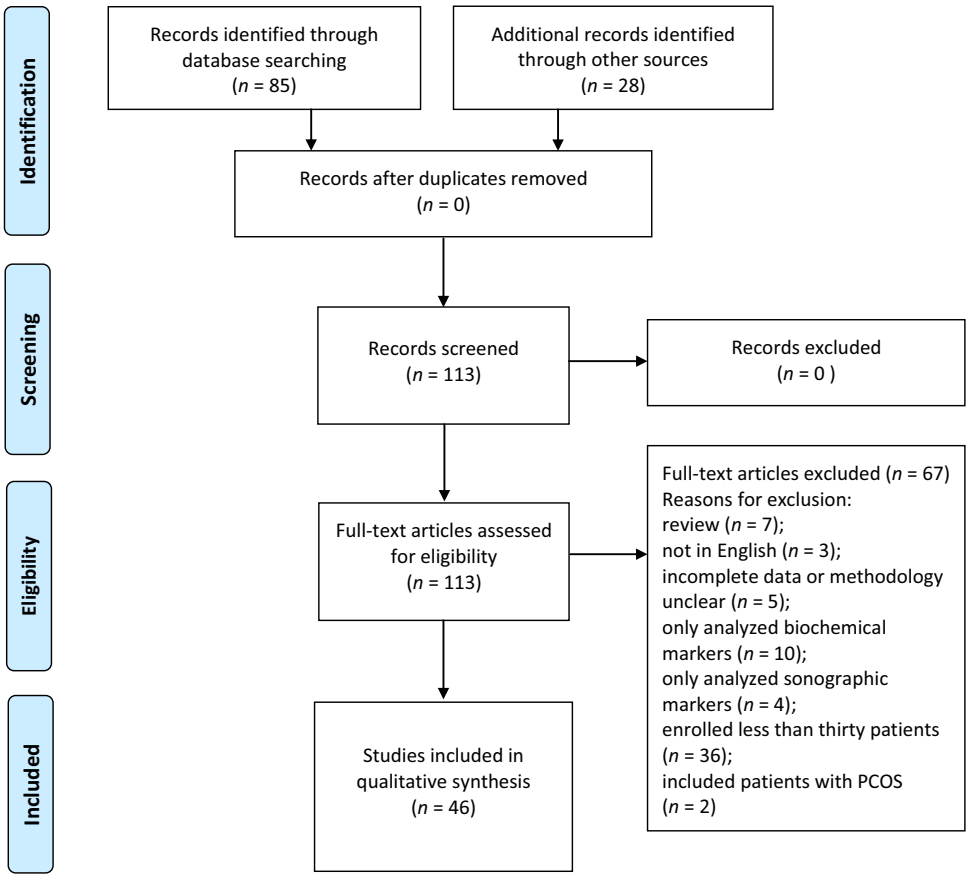

Figure 1 Flow diagram showing the selection of articles for inclusion in the review.

Table 1 Scoring system used to assess ovarian activity (adapted from Hoogland and Skouby, 1993).

Score	*Activity*	*Size of FLS (mm)*	*Oestradiol in serum (nmol/l)*	*Progesterone in serum (nmol/l)*
1	No activity	≤10	–	–
2	Potential activity	>10	–	–
3	Non-active FLS	>13	≤0.1	–
4	Active FLS	>13	>0.1	≤5
5	LUF	>13, persisting	>0.1	>5
6	Ovulation	>13, ruptured	>0.1	>5

FLS = follicle-like structure; LUF = luteinized unruptured follicle.

like structures (FLS) at ultrasound scans and measured oestradiol and *P*-values creating a score (**Table 1**). Oestradiol values were <0.1 nmol/l in women where no FLS were retrieved. The authors concluded that ultrasound analysis offers an adequate evaluation of residual ovarian activity and its degree could be considered a parameter of COC efficacy (Hoogland and Skouby, 1993).

In 2010, Spona et al. studied 40 women that used a COC containing 20 μg EE + 2 μg chlormadinone acetate for three cycles given in a 24/4-day regimen. FLS size, hormone concentrations (oestradiol, progesterone), cervical reaction and endometrial thickness were analysed. The degree of ovarian activity was assessed with the Hoogland and Skouby (H/S) score (**Table 1**). The authors did not find ovarian activity in 75% of medication cycles, but they observed residual ovarian activity in 15.9% and development of a luteinized unruptured follicle in 1.1% of COC cycles. Endometrial thickness was reduced in medication cycles to 4–5 mm compared with 10–12 mm without steroid contraception. Finally, in medication cycles, oestradiol and progesterone were lower than without medication (Spona et al., 2010).

Some authors quantified the effect of hormonal contraception on both endocrine and sonographic ovarian reserve markers in 228 women using hormonal contraception and 504 non-users. Among users, 217 (95.2%) received COC and 11 (4.8%) received a contraceptive vaginal ring. Among the users of COC, 101(46.5%) took monophasic preparations with 20 μg EE, 96 (44.2%) took monophasic preparations with 30–35 μg

EE and 20 (9.2%) used biphasic/triphasic COC. On day 2–5 of the menstrual cycle or during withdrawal bleeding, blood sampling and transvaginal sonography was performed. After adjusting for age, ovarian reserve parameters were lower among users than among non-users of hormonal contraception, in particular serum AMH concentration by 29.8%, AFC by 30.4% and ovarian volume by 42.2%. AFC in all follicle diameter categories (small, 2–4 mm; intermediate, 5–7 mm; large, 8–10 mm) was lower in users than in non-users of hormonal contraception. A negative linear association was identified between the duration of hormonal-contraception use and ovarian reserve parameters. There was not a dose–response relationship between the dose of EE and AMH or AFC. This study indicates that ovarian reserve markers are reduced in women taking hormonal contraception. Therefore, it is believed that in these women AMH concentration and AFC may not be accurate indicators of ovarian reserve (Bentzen et al., 2012).

Ovarian function and steroid dose and administration schemes

When COC were first introduced during the 1960s, a regimen based on 21 days of hormone treatment, followed by a 7-day HFI was created to mimic women's natural monthly menstrual cycles in order to achieve greater compliance and acceptability; hence it was not necessarily based on scientific evidence. Nevertheless, many authors indicate that the reduction of HFI or hormonal supplementation during HFI are beneficial in COC users as they result in a greater suppression of ovarian function (Kroll et al., 2015; Schlaff et al., 2004; Vandever et al., 2008).

Different COC formulation

There are two prospective (Crosignani et al., 1996; Jokubkiene et al., 2012) and seven randomized studies (Baerwald et al., 2004; Fitzgerald et al., 1994; Grimes et al., 1994; Rossmanith et al., 1997; Van der Does et al., 1995; Van Heusden and Fauser, 1999; Young et al., 1992) evaluating the ovarian function in women using different COC formulations.

Jokubkiene et al. estimated with three-dimensional (3D) ultrasound the ovarian volume, the number and volume of antral follicles, and with power Doppler, vascular indices such as vascularization index (VI), flow index (FI) and vascularization flow index (VFI), in women taking COC. Two hundred and ten women were included, in particular 151 were on monophasic COC and 59 on triphasic COC. One hundred and ten women were using COC with classic progestins (LNG and norethisterone [NET]) and 100 were using progestins (desogestrel [DSG], GSD, norgestimate [NGM], drospirenone and cyproterone acetate [CPA]); the oestrogen dose used was 20 μg or 30–35 μg EE. The authors detected significant variations between the different types of COC, as regards ovarian volume, number of antral follicles, size of the largest follicle, total follicular volume and vascular indices. Younger women (20–29 years old) had a significantly higher number of antral follicles measuring from 2.0–10.0 mm bilaterally, the right ovary was larger and contained more antral follicles than the left one compared with older women (30–39 years old). In all women with follicles ≤10 mm, the sonographic markers ranged without a clear difference among groups as follows: ovarian volume from 1–16 cm^3, the total follicular volume from 0.03–2.7 cm^3, VI from 0.0–13.4%, FI from 0–38, and VFI from 0.0–4.7. There was no significant variation in the proportion of women with follicle(s) >10.0 mm between the different COC formulations. The authors also showed that 11% of subjects using COC for three or more cycles had follicles >10 mm in at least one ovary on cycle day 4–8. These conclusions show that there is a great variability in sonographic markers (2D, 3D or Doppler) in women using COC and offer new information of normal findings in the ovaries of these women (Jokubkiene et al., 2012).

A randomized study assessed the ovarian follicular development in 36 women receiving one of these three COC regimes as follows: 35 μg EE for days 1–21 with 180 μg NGM for days 1–7, 215 μg NGM for days 8–14 and 250 μg NGM for days 15–21; 30 μg EE + 150 μg DSG for 21 days; 20 μg EE + 100 μg LNG for 21 days for 3 consecutive cycles of 21 days of hormonal treatment followed by 7 days of HFI. The follicular development was evaluated by ultrasound scan every third day. Follicles >10 mm were found in 16 of 36 women (44%) and follicles >14 mm were found in 9 of the 17 women (53%) who showed higher EE concentrations (mean maximum concentration of 630.6 ± 112.5 pmol/ml). Eighty-six percent of follicles >10 mm grew during HFI. No ovulations were documented. These results suggest that follicular growth to a diameter comparable to that of ovulatory follicles in natural cycles is observed during compliant COC use and is associated with initial recovery of endocrine activity that usually can occur during the HFI (Baerwald et al., 2004).

Another study evaluated the pituitary-ovarian recovery that emerged during HFI in women using three different low-dose COC. Hormone concentrations (LH, FSH, oestradiol) were determined and daily ultrasound scans were performed to estimate follicle number and size in 44 healthy women using one of the following three regimes: 20 μg EE + 75 μg GSD, 20 μg EE + 150 μg DSG or 30 μg EE + 150 μg DSG. No ovulations were observed. At the beginning of HFI, hormones concentrations were not statistically significantly different between the study groups. FSH concentrations were significantly greater at the end of the HFI in the 30 μg EE group compared with both 20 μg EE groups ($P = 0.001$). Follicular size was significantly smaller at the beginning and at the end of HFI in the 30 μg EE group compared with both 20 μg EE study groups. Dominant follicles (diameter >10 mm) were retrieved at the end of HFI only in the 20 μg EE study groups (in 27% of women taking 20 μg EE + 75 μg GSD and 18% of women taking 20 μg EE + 150 μg DSG, respectively). Finally, the area-under-the-curve for oestradiol was statistically significantly lower in the 30 μg EE group compared with both 20 μg EE groups. These results prove that the degree of residual ovarian activity at the beginning of HFI is determined by EE dose rather than the progestin component of COC (Van Heusden and Fauser, 1999).

Rossmanith et al. enrolled 118 women to evaluate the inhibition of ovarian activity and anticonceptive action on the cervix and endometrium during use of two low-dose monophasic COC for three treatment cycles: 20 μg EE, 500 μg NET (group a); 20 μg EE, 150 μg DSG (group b). Hormone concentrations (LH, FSH, oestradiol and progesterone) were measured and simultaneous ultrasound evaluations (to determine follicular development, cysts and endometrial thickness) were

performed to examine ovarian activity. No ovarian activity was observed in most of the treatment cycles (90.8% of group a and 97.2% of group b, respectively). Follicular activity or cyst development were found only in a few of the investigated cycles (10.4% of group a and 2.8% of group b, respectively). Gonadotrophin concentrations were suppressed in most treatment cycles (group a: 76.6% versus group b: 84.8%). Follicular activity was detected in 19.3% (group a) versus 12.2% (group b) of all cycles, in association with oestradiol serum concentrations >0.1 nmol/l. No ovulations were observed, combining ultrasound parameters and hormone concentrations, in any treatment cycle, however, in 4.1% (group a) versus 2.9% (group b) of investigated cycles, serum progesterone concentrations exceeded 5 nmol/l, indicating that ovulation had presumably occurred. Endometrial thickness and cervical mucus quantity and quality were suppressed during most pill cycles. In conclusion ovarian activity seems to be inhibited in the majority of cycles in women using low-dose contraceptives (Rossmanith et al., 1997).

Others authors enrolled 51 women taking COC, to evaluate ovarian activity by means of ultrasound monitoring of follicular growth and serum hormone concentrations (oestradiol and progesterone). Twenty-two women used a triphasic COC containing 35 μg EE + 50 μg DSG in the first seven tablets; 30 μg EE + 100 μg DSG in tablets 8–14, and 30 μg EE + 150 μg DSG in tablets 15–22; 29 women took one of the two COC containing 20 μg EE; 20 μg EE + 150 μg DSG (15 women) or 20 μg EE + 75 μg GSD (14 women). Women received COC for 86 treatment cycles. Follicular growth was detected in nine patients with similar frequency during the 3rd or 4th cycle (9%) and during the 6th, 7th or 8th cycle (11%). Hormone concentrations were suppressed in any cycle but there was no correlation between FLS and oestradiol and progesterone serum concentrations. These observations suggest that, even with low-dose COC use, residual ovarian activity could persist without ovulation (Crosignani et al., 1996).

In a randomized trial of 1995, Van Der Does et al. evaluated the presence of ovulation, assessed combining by transvaginal ultrasound evaluation and serum oestradiol, and progesterone determinations in 31 women using two triphasic COC: the first containing 30 μg EE + 50 μg LNG (days 1–6), 40 μg EE + 75 μg LNG (days 7–11) and 30 μg EE + 125 μg LNG (days 12–21), the second containing 35 μg EE + 50 μg DSG (days 1–7), 30 μg EE + 100 μg DSG (days 8–14) and 30 μg EE + 150 μg DSG (days 15–21), during six cycles of hormonal therapy. No evidence of ovarian activity was reported in 10 subjects. The remaining 21 women grew follicles during HFI but FLS were reduced in volume or disappeared in the first pill week. One woman using triphasic DSG showed a luteinized unruptured follicle and in one woman receiving triphasic LNG ovulation presumably had occurred. The two triphasic COC inhibited ovarian activity at the same level. Ovarian activity tended to increase with prolonged COC use in both treatment groups (Van der Does et al., 1995).

Grimes et al. estimated the risk of follicular development and ovulation in 40 women randomized to three different pill regimens (a triphasic pill containing 0.5 μg NET + 35 μg EE on days 1–7, 0.75 μg + 35 μg on days 8–14 and 1 μg + 35 μg on days 15–21, followed by 7 days of inert tablets; a monophasic pill containing 1.0 μg NET + 35 μg EE for 21 days followed by 7 days of inert tablets; and a monophasic pill containing 0.5 μg NET + 35 μg EE for 21 days followed by 7 days of inert tablets) or to non-steroidal contraception. Vaginal ultrasonography was performed to evaluate follicular development and serum progesterone concentrations were measured to establish if ovulation occurred. Women using the higher-dose monophasic pill had an increased likelihood to develop a FLS >30 mm during a treatment cycle (relative risk [RR] 0.5). Women taking a monophasic or multiphasic pill had a comparable risk to grow FLS (RR 1.3). With the multiphasic pill the maximum ovulation rate over 60 cycles was 1.7 per 100 cycles. In conclusion, the lower-dose rather than the higher-dose monophasic pill showed the same effectiveness of the multiphasic pill in its suppression of follicular development (Grimes et al., 1994).

Young et al. compared the incidence, risk, size and time to resolution of ovarian follicles, evaluated with ultrasound scans in healthy women who took lower-dose and triphasic COC or a placebo. Forty-eight patients were randomized to use a monophasic pill with 30 μg EE + 1.5 μg NET acetate taken for 21 days or a triphasic pill containing the same progestin dosage with 20, 3, and 35 μg EE taken on days 1–5, 6–12 and 13–21, respectively. Sixty-three percent of placebo-treated subjects developed follicles >18 mm, compared with 39% and 23% in the triphasic and monophasic pill groups. The risk for each group of developing a large follicle during a single cycle was not different. No dominant follicle persisted for >2 weeks for any subject. In conclusion, the follicular development continues during treatment with COC. In addition, the findings fail to support the hypothesis that triphasic COC result in persistent ovarian cysts (Young et al., 1992).

Other authors compared in a randomized study the influence of two low-dose monophasic COC on the suppression of ovulation by means of hormonal concentrations (oestradiol and progesterone) determination and transvaginal ultrasound evaluation. They enrolled 52 women receiving one of two low-dose monophasic COC (20 μg EE + 75 μg GSD or 20 μg EE + 150 μg DSG) for three treatment cycles. No ovulations were observed in any COC group. Some FLS in association with serum oestradiol concentration increase were described in 21% of women in at least one treatment cycle. The two combined COC showed no significant differences on the residual ovarian function. Hormone concentrations were significantly decreased throughout all three treatment cycles. Mean LH and FSH concentrations were similar with both preparations. (Fitzgerald et al., 1994).

COC versus vaginal ring

In a randomized open-label trial, Duijkers et al. compared the action of the combined contraceptive vaginal ring and a COC on ovarian function. Forty women were randomly allocated to receive the vaginal ring (NuvaRing, 21 subjects) or a COC (30 μg EE + 150 μg LNG, 19 subjects) for two months. The vaginal ring was initiated on cycle day 5, COC on cycle day 1. Ultrasound parameters (follicular size and endometrial thickness) and hormone serum concentrations (FSH, LH, oestradiol and progesterone) were analysed. The median maximum follicular diameter (maxFD) was ≤11 mm during hormonal therapy. Women using the vaginal ring showed more follicles than women taking a COC, in the first treatment cycle, because of the different onset of preparations. MaxFD were not different in the second treatment cycle. In both groups,

oestradiol and progesterone concentrations remained low during hormonal therapy. No ovulations were described. Both preparations showed a similar effect on the suppression of ovarian activity. In the first treatment cycle, for the different onset of the two contraceptives, ovarian activity was lower in the COC group. In the second cycle, ovarian inhibition was comparable between the two study groups (Duijkers et al., 2004).

Transdermal patch

In an open-label study in 2004, Heger-Mahn et al. enrolled 199 healthy women, aged between 18 and 35 years, to determine the effectiveness in the inhibition of ovulation of a transdermal, combined hormonal contraceptive patch containing 0.9 mg EE+ 1.9 mg GSD. Women used one patch per week for 3 weeks, followed by 1 week of no treatment, for two months. The H/S score was used to determine ovarian activity by means of transvaginal ultrasonography and serum hormone measurements (oestradiol, progesterone and LH). No ovulation was observed in all participants. A return to ovulation was observed in 85.7% of women during the first month after the ending of hormonal therapy (Heger-Mahn et al., 2004).

Modification of the HFI

Three randomized studies (Killick et al., 1998; Rible et al., 2009; Spona et al., 1996) and one prospective study (Sullivan et al., 1999) evaluated the suppression of ovarian activity after HFI modifications.

In 1996, Spona et al. performed a double-blind study of 60 multicentre randomized women, based on the administration of 20 μg EE +75 μg GSD for 21 or 23 days to assess the suppressive action on ovarian activity. Diameters of FLS were calculated by ultrasound scans. By means of ultrasound study (to evaluate follicular growth) and determination of LH, FSH, oestradiol and progesterone serum concentrations, they noticed a suppression of ovarian activity most pronounced in the 23-day scheme. Thus, they came to the conclusion that shortening the HFI in the low-dose COC regime may increase the margin of safety in women. Indeed, the 23-days formulation adequately suppresses the ovulation, allows a good cycle control and does not increase the incidence of side effects (Spona et al., 1996).

In 1998, a prospective, randomized, double-blind trial included 47 women to assess the effect of low EE doses, given during the nominal HFI, on the ovarian activity. They administered 20 μg EE + 150 μg DSG for 21 days followed by either a placebo for 7 days or a placebo for 2 days and 10 μg EE for 5 days. During each treatment cycle, ultrasound scans and hormone concentration determinations (oestradiol and progesterone) were performed to evaluate ovarian activity. Women taking 10 μg EE per day during the last 5 days of the 7-day nominal HFI showed a bigger inhibition of ovarian activity and reduced folliculogenesis than provided by the placebo in women using the same 150 μg DSG +20 μg EE regimen during the first 21 days of each cycle. It is shown that the administration of EE during the HFI, more effectively suppresses ovarian follicular activity (Killick et al., 1998).

In 1999, a prospective cohort study enrolled 58 women, who were given 15 μg EE + 60 μg GSD for 21 or 24 days, to compare the ovulation suppression and the ovarian function. Ultrasonographic data and serum hormone concentrations (oestradiol, progesterone, FSH and LH) were determined to measure ovarian activity. Ovulation occurred when a FLS of 13 mm in size was detected and its rupture occurred within 48 h in association with serum oestradiol and progesterone concentrations of 0.03 ng/ml and 1.6 ng/ml, respectively, in the same cycle. They observed that women taking a COC with 15 μg EE + 60 μg GSD in a 24-day regimen, showed greater ovarian suppression with reduced determination of FLS and lower serum oestradiol concentrations (Sullivan et al., 1999).

In 2009, other authors evaluated 41 women in an open-label trial, with the aim of documenting the differences in follicular growth during a 7-day versus 4-day HFI in a COC scheme with 20 μg EE + 1 mg NET acetate for three 28-day pill cycles. Ovarian activity was evaluated by means of hormone concentration assays (FSH, LH, oestradiol, progesterone and inhibin B concentrations) and ultrasound scan evaluating ovarian follicular growth and endometrial thickness. Follicle size of the largest visible FLS was measured and a H/S score was calculated. It was shown that administering a COC with 20 μg EE + 1 mg NET acetate for 21 days with 7-day HFI or for 24 days with 4-day HFI, there was no significant difference in follicular growth and ovarian steroid activity in two different regimens. This result indicates that the benefits of a shortened HFI may be altered by the progestin formulation used (Rible et al., 2009).

Continuous versus cyclical contraception

Ovarian function during continuous versus cyclical contraception was evaluated by one prospective study (Archer et al., 2009) and two randomized studies (Birtch et al., 2006; Legro et al., 2008).

In 2006, Birtch et al. enrolled 36 women in a randomized study to evaluate the ovarian follicular growth during cyclic versus continuous COC dosing regimens, in order to determine the follicular growth during the HFI and following COC discontinuation. Each woman received one of two different monophasic COC formulations (30 μg EE + 150 μg LNG or 35 μg EE + 250 μg NGM) for three 28-day cycles according to either a conventional (21 days) or continuous regimen. Transvaginal ultrasonography was executed to examine ovarian follicular development. Progesterone and oestradiol concentrations were measured. They showed that for both formulations more dominant follicles developed during conventional oral contraceptive than continuous oral contraceptive use (8 versus 0, respectively; $P = 0.01$). The continuous regimen resulted, therefore, more effective in preventing ovulation (Birtch et al., 2006).

A prospective randomized double-blind trial of 62 patients determined the effect of continuous versus cyclical oral contraception on the ovarian function. Women were given 20 μg EE + 1 mg NET acetate for 6 cycles of 28 days each, in the form of continuous regimen or with a pause of 7 days. Ultrasound scans for ovarian size and endometrial thickness were performed and serum concentrations of sex steroids, gonadotrophins, insulin, glucose, lipid profile, sex hormone-binding globulin (SHBG) and urinary concentrations of estrone

3-glucuronide (E1G) and pregnanediol 3-glucuronide (PdG) were assayed. Women in the continuous group showed a significant reduction of serum oestradiol and integrated urinary oestrogens from baseline and had smaller ovaries and dominant follicles, demonstrating that the continuous regimen is associated with greater suppression of ovarian function (Legro et al., 2008).

Another prospective study enrolled 37 patients using a continuous COC, namely oral 90 μg LNG + 20 μg EE, to evaluate the suppression of ovulation and time to return to ovulation after interrupting hormonal therapy. Ultrasound scans and serum hormone measurements (oestradiol, progesterone, FSH and LH) were performed to characterize ovarian activity. The continuous LNG + EE regimen (28 days for 3 cycles) completely suppressed ovulation, with poor follicular growth documented by ultrasound and with fast recovery of ovulation after interrupting COC use (Archer et al., 2009).

Supplementation of the HFI with EE

Three randomized studies (Schlaff et al., 2004; Seidman et al., 2015; Vandever et al., 2008) and one prospective study (Kroll et al., 2015) evaluated the suppression of ovarian function after the supplementation of EE during HFI.

In 2004, Schlaff et al. randomly assigned 54 women to receive one of these three different regimens: either 20 μg EE + 100 μg LNG followed by seven pill-free days, or 20 μg EE + 150 μg DSG followed by 2 days of placebo then 10 μg EE for 5 days, or 28 days of 20 μg EE + 150 μg DSG. Ovarian inhibition was assessed by ultrasonographic evaluations and by measurements of serum gonadotrophins, and daily urinary conjugates of oestrogen and progesterone for two treatment cycles. They demonstrated that women taking a contraceptive pill with a 7-day HFI showed an increased follicular development compared with women who took a supplementation with either oestrogen alone or oestrogen plus progestin (Schlaff et al., 2004).

Other authors led a prospective randomized study to estimate follicular growth and hormone concentrations with three COC regimens before, during and after the 7-day HFI or 7-day EE-supplemented interval. Thirty-three women were enrolled and treated with a COC in the standard 21/7 regimen for at least 2 months. Following that they were allocated to treatment with 30 μg EE + 150 μg LNG according to one of three different patterns: 150 μg LNG +30 μg EE for 21 days followed by 7 days of placebo or 150 μg LNG + 30 μg EE for 84 days followed by 7 days of placebo or 150 μg LNG + 30 μg EE for 84 days followed by 7 days of 10 μg EE. To evaluate the degree of ovarian activity ultrasound scans were performed and FSH, LH, oestradiol and inhibin B concentration were measured. They showed that supplementation of the standard 7-day HFI with 10 μg EE after 84 days of an extended COC reduced both FSH concentrations and the number of growing follicles (Vandever et al., 2008).

In a randomized, open-label trial, Seidman et al. described ovarian activity inhibition during a 21/7 day active low-dose COC scheme that included only EE during the traditional HFI (150 μg DSG + 20 μg EE for 21 days + 7 days 10 μg EE) and two 28-day regimens, a 24/4 day regimen of 3 mg drospirenone (DRSP) + 20 μg EE 24 days + 4 days placebo and a 21/7 regimen of 100 μg LNG + 20 μg EE for 21 days + 7 days placebo. Ovarian activity suppression was examined by ultrasound scans and serum hormone measurements (progesterone, oestradiol, FSH and LH) and assessed using the H/S score. All three regimens showed a low ovarian activity rate (H/S grade 4 or 5), in particular 0% for 21 days DSG + EE + 7 days EE, 1% for 24 days DRSP + EE + 4 days placebo and 1% for 21 days LNG + EE + 7 days placebo. A similar suppression of serum hormone concentration was present in all three treatments. The 21/7-active low-dose COC regimen (21 days DSG + EE + 7 days EE), that included only EE during the traditional HFI, showed the similar reduction of follicular development that was observed during the 24 days DRSP + EE + 4 days placebo and the LNG + EE + 7 days placebo regimens (Seidman et al., 2015).

Recently, Kroll et al. evaluated the role of a 91-day extended COC regimen (84 days of 150 μg LNG + 30 μg EE plus 7 days of 10 μg EE) on ovarian activity. Follicular development was quantified using the H/S score. No luteinized, unruptured follicles or ovulation were detected in the 35 subjects included in the efficacy analysis during the first 28-day interval; subsequently ovarian activity was detected in 1 of 35 women (2.9%) in the second 28-day interval; and in 2 of 35 women (5.7%) in the final 35-day interval. Overall, the ovarian activity rate was 2.9% for the 91-day treatment period. The authors concluded that the 91-day extended-regimen COC with low-dose EE supplementation has shown to effectively inhibit ovarian activity and suppress ovulation and women showed good compliance (Kroll et al., 2015).

Progestogen-only pill (POP)

Natural progesterone or synthetic progestins contained in most COC show a very low influence over FSH secretion. Follicular growth continues during administration of POP and, indeed, in some low dose regimens, ovulation is not always inhibited (Landgren and Diczfalusy, 1980). In fact, ovulation can occur in 30–40% of POP users (McCann and Potter, 1994). Also data obtained from progestin implants indicate a variable degree of ovarian activity ranging from normal ovulatory cycles to total suppression of follicle development (Mäkäräinen et al., 1998). Only one randomized study evaluated the effect of a POP on ovarian function (Duijkers et al., 2015). This randomized trial compared the effect on follicular development of a POP containing 75 μg DSG and a new POP containing 4 mg DRSP in a 24/4-day regimen. Follicular diameter, serum oestradiol (oestradiol) and progesterone concentrations were measured to determine H/S scores. Both treatments effectively suppressed ovulation, which was demonstrated by the similar follicular diameters, oestradiol concentrations and H/S scores. The authors concluded that the new DRSP-only pill was as effective as the DSG-only pill in inhibiting ovulation.

Subdermal contraceptive implants

Three prospective studies evaluated ovarian activity in women using subdermal contraceptive implants (Alvarez-Sanchez et al., 2000; Brache et al., 2000; Shaaban et al., 1993).

Shaaban et al. studied the probability of ovulation and subclinical abortion in 50 women who were using a LNG implant

(Norplant) for >1 year compared with 35 ovulatory cycles in normal fertile women not using contraception. Sonographic and hormonal evidence of ovulation were observed in one third of LNG implant users; two of them resulted in conception. However, the majority of these ovulatory cycles showed low midcycle peaks of oestradiol, FSH and LH, and evidence of luteal phase defect (LPD). LNG implant users had also significantly thinner endometrium that did not exhibit the normal phasic changes in sonographic texture. Furthermore, excessive follicular enlargement was observed in 46% of the cycles of LNG implant users (Shaaban et al., 1993).

Other authors evaluated the ovarian activity in women using Nestorone progestin (NES). NES is a potent 19-norprogesterone, which is active only via subdermal implants. This study compared sonographic and endocrine parameters during use of either one 4 cm or two 3 cm NES implants for 24 months. Sixty participants were included in each dose group. Follicular development was not fully suppressed during use of either NES implant regimens, and serum oestradiol concentrations continuously <100 pmol/l were infrequently observed. Women using the two-implant system showed stronger suppression of ovarian activity, which was less than 5% during 18 months of use, compared with those using the one-implant system, who showed suppression of approximately 27%. On the other hand, a single implant is easier to insert and remove (Brache et al., 2000).

In 2000, Alvarez-Sanchez et al. enrolled 103 users of subdermal LNG implants (Norplant) and 50 users of the TCu380A intrauterine contraceptive device (IUD), to study with vaginal ultrasonographic assay the presence of enlarged follicles. A single blood sample for oestradiol and progesterone concentration measurements was taken at the time of enrolment and was repeated weekly in the women with enlarged follicles who were being followed up until follicle involution. Follicles greater than 25 mm were revealed by ultrasonography in 17.5% of LNG implant users and 4% of TCu380A IUD users, respectively ($P < 0.04$). Progesterone concentrations were low in all LNG implant users with enlarged follicles and were 17.4 and 8.7 nmol/l in the two users of the TCu380A IUD who had enlarged follicles. At most, 4 weeks were necessary for the involution of enlarged follicles (Alvarez-Sanchez et al., 2000).

Ovarian function following missed or delayed doses of hormonal contraceptives

Hormonal contraception is very effective if correctly taken, whilst incorrect use is the major cause of failure of the contraceptive. Five randomized studies and two prospective studies evaluated the effect of a missing or delayed dose of hormonal contraceptive on ovarian function (Brache et al., 1999; Creinin et al., 2002; Elomaa et al., 1998; Mulders et al., 2002; Petta et al., 1998, 2001; Pierson et al., 2003).

In a randomized study Creinin et al. evaluated follicular development in 79 patients, using COC containing triphasic NGM + EE ($n = 40$) or monophasic LNG + EE ($n = 39$), with intentional imperfect compliance, for two months of treatment. One group received a 35 μg EE preparation with NGM 180 μg (days 1–7), 215 μg (days 8–14) and 250 μg (days 15–21). The second group received a 20 μg EE formulation with LNG 100 μg (days 1–21). After completing a 28-day cycle, women were instructed to extend the pill-free interval from 7 days to 9 days by missing on purpose the first two active pills of the following blister pack. Women using LNG + EE developed follicles with a mean maximum diameter that was significantly greater compared with women using NGM + EE ($P = 0.047$). Median serum oestradiol concentrations were significantly greater in women using LNG + EE compared with those using NGM + EE on pill days 10 ($P < 0.001$) and 14 ($P = 0.001$). Progesterone concentration ≥3 ng/ml (considered consistent with presumptive ovulation) was observed in two women in the NGM + EE group and in three women in the LNG + EE group; nevertheless, follicles bigger than 13 mm were not observed in these women. Women taking LNG + EE had significantly greater follicular activity after a longer HFI compared with those taking triphasic NGM + EE. The authors stated that although follicular development was significantly increased among LNG + EE users as compared with NGM + EE users following an extended pill-free interval, further studies are required to assess the clinical importance of this result (Creinin et al., 2002).

Another study investigated whether missing the first three tablets of the hormone treatment scheme results in ovulation by extending further the pill-free period from 7–10 days. Ninety-nine women were randomized to receive one of three contraceptive regimens: monophasic GSD consisting of 75 μg GSD + 30 μg EE; triphasic GSD, consisting of 6 days of 50 μg GSD + 30 μg EE, 5 days of 70 μg GSD + 40 μg EE and 10 days of 100 μg GSD + 30 μg EE; monophasic DSG consisting of 150 μg DSG + 20 μg EE. Pituitary-ovarian function was assessed by ultrasound scans and determination of hormone concentrations (oestradiol, progesterone and FSH). No ovulations were described. However, FSH and oestradiol concentrations were higher during the first seven pill-free days. After 10 pill-free days, follicles with diameters >18 mm were detected in 24%, 24% and 40% of the monophasic GSD, triphasic GSD and monophasic DSG groups, respectively. Finally, with missing doses from the first one to three pills of a medication cycle, preovulatory follicles were frequently found, but without ovulation occurrence (Elomaa et al., 1998).

In a prospective randomized trial, 158 women were enrolled and administered 25 μg depot medroxyprogesterone acetate (DMPA) + 5 μg oestradiol cypionate (oestradiol C) with injection on day 5 or on day 7 of their menstrual cycle. Progesterone serum concentrations were measured and vaginal ultrasound examinations were performed to evaluate ovarian activity. In women who received DMPA + oestradiol C on day 5 only follicles <16 mm were detected. Among those women who received DMPA + oestradiol C on day 7, 18% exhibited follicles >16 mm and in 3% of this subgroup ovulation occurred. Inhibition of ovarian activity was higher when the injection of DMPA + oestradiol C was administered on day 5 rather than on day 7 (Petta et al., 2001).

In 2002, Mulders et al. carried out an open-label randomized trial to evaluate ovarian activity in 45 women who used an etonogestrel/ethinyl oestradiol vaginal ring (NuvaRing), who were trained to follow, or not, the recommended scheme. All women used the ring for one cycle according to the recommended regimen. Women in group A ($n = 15$) continued with a 'normal' 3-week period of ring use. Following this their ovarian function was monitored by daily vaginal ultrasound and serum hormone concentrations. Women in group B ($n = 15$), continued with three consecutive days of ring use only, after which each woman was observed until ovulation. Women

in group C ($n = 15$) were not allowed to start a second 'normal' cycle until a follicle with a diameter of 13 mm was identified by ultrasound; subsequently, these participants started another 'normal' cycle and follicular growth was monitored daily. Despite the difference in second cycle length in group A (3 weeks) and group B (3 days), a new cohort of follicles developed in both groups and the time to ovulation after ring removal was similar (19 versus 17 days). In group C the median time to development of a follicle up to 13 mm in size was 11 days (range 8–21 days); none of the women in this group ovulated after insertion of the second ring. The authors showed that the etonogestrel/ethinyl oestradiol vaginal ring is a highly effective, reversible method of hormonal contraception (Mulders et al., 2002).

In a randomized open-label study the contraceptive patch was compared with COC by maximum mean follicular diameter and ovulation rate in normal cycles and after dosing errors. The participants ($n = 184$) received either the 20-cm^2 patch designed to deliver 150 μg norelgestromin + 20 μg EE daily to systemic circulation, (groups 1 and 2), or one of three COC (a triphasic LNG COC containing 50 μg LNG + 30 μg EE for days 1–6, 75 μg LNG + 40 μg EE for days 7–11, 125 μg LNG 125 +30 μg EE 30 for days 12–21 and placebo for days 22–28; a monophasic LNG COC containing 100 μg LNG + 20 μg EE for days 1–21, a placebo for days 22–28; or a triphasic NGM COC containing 180 μg NGM + 35 μg EE for days 1–7, 215 μg NGM + 35 μg EE for days 8–14, 250 μg NGM + 35 μg EE for days 15–21 and placebo for days 22–28). Correct doses were used in cycles 1, 2, 3 and 5 and dosing errors were planned for cycle 4, to give a shortened 10-day cycle. For cycle 4 in patch group 1 women wore one patch for the first 10 consecutive days. In patch group 2 and the COC groups, the first seven dosing days were followed by three drug-free days. After a 3-day dosing error, follicular diameter was significantly smaller in the patch group (mean, 7.0 mm) versus each COC group (range of means, 11.8–17.1 mm). Similar results were seen after correct dosing (group 1). Patch users demonstrated a significantly lower incidence of ovulation compared with women using COC. Follicular diameter and ovulation rate were significantly reduced among patch users compared with COC users in normal cycles and after dosing errors (Pierson et al., 2003).

Brache et al. designed a prospective study to determine the timing of onset of contraceptive effectiveness in LNG implant (Norplant) users when inserted in days 8–13 of the cycle. Serum samples of oestradiol, progesterone, LH and LNG were measured. Ovulation, as defined by progesterone >2.5 ng/ml, occurred in 40% of participants. Therefore, anovulatory cycles occurred in the remaining 60% of participants with two distinct oestradiol profiles: continuously increasing oestradiol concentrations to a high mean of 0.41 ng/ml (28%), or no sustained increase in oestradiol (32%). Since ovulation will either occur within 48 h of insertion of the implant or will be impaired, additional contraceptive protection is required for 3 days only (Brache et al., 1999).

Petta et al. tried to determine by means of a prospective study the timing of onset of contraceptive effectiveness after the first injection of 150 μg of depot medroxyprogesterone acetate (DMPA) administered in 30 women between days 8 and 13 of the menstrual cycle. Ovarian function was assessed by serum concentrations of oestradiol and progesterone and ultrasound scans monitoring follicular development. All the ovulations occurred in nine (30%) of 30 women receiving DMPA between days 10 and 13 of the cycle. No woman who received injections on day 8 or 9 ovulated. Women with low ovarian activity showed a greater suppression of ovulation. All the ovulations occurred within three days after the injection. Therefore, an additional contraceptive method is recommended until seven days after the first injection of DMPA (Petta et al., 1998).

Follicle cysts and oral contraceptive use

It has been observed that women using POP showed an increased tendency to develop functional ovarian cysts (Tayob et al., 1985), probably related to the reduced impact of progestins on FSH secretion. In literature it is also documented that there is a greater number of follicle cysts in women using multiphasic and low-dose monophasic combined COC than in those using moderate-dose monophasic COC (Lanes et al., 1992). It is still not clear the mechanism underlying the growth of follicle cysts during combined COC (Baerwald and Pierson, 2004). However, it is possible that low-dose COC do not protect against functional cysts formation to the same extent as high-dose formulations and it could be related to lower hypothalamo-pituitary ovarian suppression.

Two randomized (Broome et al., 1995; Egarter et al., 1995), one cohort (Lanes et al., 1992) and two case-control studies (Holt et al., 1992, 2003) evaluated the risk of functional ovarian cyst development during COC use.

Egarter et al. investigated in a randomized study the presence of ovarian follicles and cysts with ultrasound evaluation and determined serum hormone concentrations in 65 women receiving either 20 μg EE + 150 μg DSG (group A) or 35 μg EE + 250 μg NGM (group B) for two treatment cycles. Before starting hormonal therapy, at least one follicle <35 mm in diameter was observed in 39% of women in group A and 31% in group B. By the end of the 2-month study period, follicles <35 mm were detected in only 14% of women in each group. An ovarian cyst >35 mm was found in only one subject who received the 35 μg EE preparation. Ovulation, identified by hormone concentrations, was documented in one subject in each group; no pregnancy occurred in either group. Women receiving lower dose oestrogen COC did not show an increased incidence of ovarian follicles or cysts compared with women using a 35 μg EE preparation, and this could be explained by the type and dose of the progestin used (Egarter et al., 1995).

Holt et al. evaluated in 106 women the risk of functional ovarian cyst development, with surgical or ultrasound evaluation, during the use of monophasic or triphasic COC. Compared with women not using hormonal contraception, the relative risks of a diagnosed functional ovarian cyst among women currently using COC were 0.8 for users of monophasic COC and 1.3 for users of triphasic COC. The results of this study suggest that subjects currently using low-dose monophasic COC do not have a substantially decreased risk of functional ovarian cyst formation. According to this study the use of triphasic COC does not increase the risk of functional ovarian cysts (Holt et al., 1992).

In 2003, a case-control study was conducted to determine whether COC and tubal sterilization affects functional ovarian cyst risk. They enrolled women, in particular 392 cases

and 623 controls, showing at ultrasound scan or during surgery a functional ovarian cyst. Women in the case group received the following different types and doses of COC: greater than 35 μg EE monophasic, 35 μg EE monophasic, less than 35 μg EE monophasic, multiphasic (triphasic and biphasic) and POP. The purpose of the study was to evaluate the association between the diagnosis of a functional ovarian cyst and current contraceptive method. The overall odds ratio (OR) was 0.72 in women using COC, compared with women using non-surgical, non-hormonal contraception or no contraception. Women receiving 35 μg EE monophasic COC had a slightly lower risk (OR 0.69) of functional ovarian cyst compared with women receiving less than 35 μg EE monophasic (OR 0.79) or multiphasic COC (OR 0.76). In conclusion, low-dose COC is associated with a low or no influence on functional ovarian cyst probability (Holt et al., 2003).

Other authors tried to determine whether multiphasic, low-dose monophasic and high-dose monophasic COC have a similar effect in inhibiting functional ovarian cysts development. They enrolled 7462 healthy women and 32 women with evidence of a functional ovarian cyst >20 mm in diameter. COC were grouped into four types: multiphasic pills, low-dose monophasic pills (≤35 μg oestrogen), high-dose monophasic pills (>35 μg oestrogen) and POP. The incidence of functional ovarian cysts was lower among women receiving multiphasic pills (rate ratio 0.91), low-dose monophasic pills with ≤35 μg oestrogen (rate ratio 0.52) and high-dose monophasic pills with >35 μg oestrogen (rate ratio 0.24), compared with women who did not use a COC. Low-dose COC, with a smaller hormonal potency, may show a slightly lower suppression of functional ovarian cysts formation than high-dose monophasic pills whose protective action is already known in literature (Lanes et al., 1992).

Broome et al. in 1995 performed a randomized study of 17, 15 and 10 women who were administered triphasic contraceptives, progestins or nothing. The study aimed to establish whether the incidence of ovarian cyst development was different among triphasic COC users or POP users, and to evaluate the persistence of any diagnosed cysts. Women were subcategorized into three groups by the contraceptive method used. Group T comprised 17 subjects who had been taking a triphasic COC for at least six treatment cycles. This regimen consisted of 6 days of 30 μg EE + 50 μg LNG, 5 days of 40 μg EE + 75 μg LNG and 10 days of 30 μg EE + 125 μg LNG, followed by 7 tablet-free days. Group P included 15 women who had been taking a POP either 30 μg LNG or 350 μg NET daily for at least six months. Both groups T and P were studied for three treatment cycles. Group C (the control group) included 10 women who were not administered any hormonal contraceptive or an IUD for at least 6 months. They were studied like groups T and P over three months, during which they did not take hormonal contraceptives. Ultrasound evaluation was performed to count and measure follicles ranging from 10 mm to 30 mm in size. Functional ovarian cysts consisted of any fluid-filled formation greater than 30 mm in size identified in the second half of the cycle that remained for more than two cycles; an enlarged follicle was defined as any analogous structure that did not persist. It was found that women taking COC had a lower incidence of enlarged follicles compared with women in the POP group. In addition, the study showed that any swollen follicle was transitory (Broome et al., 1995).

Conclusion

The purpose of this review is to give a complete evaluation of residual ovarian activity during hormonal contraceptive use. Unfortunately, it is difficult to draw a final conclusion because the analysed studies are very heterogeneous. They evaluate different outcome parameters such as AFC, AMH concentration, ovarian volume, VI, H/S scores, hormonal assay (FSH, LH, oestradiol, progesterone, inhibin B, E1G, PdG and SHBG), endometrial thickness and development of follicle structures; moreover, every type and dosage (EP or only progestin) of hormonal contraceptive and every mode of administration (oral, vaginal ring, implant or transdermal patch) is used. Hence, it is very difficult to make a comparison of the results. Nevertheless, important evidence emerges from this analysis.

Follicular development continues during treatment with hormonal contraceptives, although each regimen of steroids inhibits ovulation, if taken correctly (Baerwald et al., 2004; Crosignani et al., 1996; Spona et al., 1996; Young et al., 1992).

During treatment with hormonal contraceptives FSH, LH and oestradiol serum concentrations decrease (Deb et al., 2012; Fitzgerald et al., 1994). There is also a reduction of the endometrial thickness (Spona et al., 2010).

Some studies evaluate ovarian reserve parameters, namely serum AMH concentration, AFC and ovarian volume among users and non-users of hormonal contraception. In particular, Bentzen et al. show that AFC in all follicle size categories (small, 24 mm; intermediate, 5–7 mm; large, 8–10 mm) is lower in users than in non-users of hormonal contraception, as well as AMH serum concentration (Bentzen et al., 2012). Deb et al. instead, do not a find significant difference in serum AMH concentrations between these two groups (Deb et al., 2012). As regards the effect of hormonal contraception on AMH, there are still controversies in literature. There are many articles about it, but none of them had been included in our review because these studies did not fit the inclusion criteria. Anyway, some authors claim that AMH concentration, both in healthy women and in those suffering from polycystic ovary syndrome (PCOS), is not influenced by COC, vaginal contraceptives, combined injectable contraceptive, POP, progestogen-only injectable, LNG intrauterine system or pregnancy (Li et al., 2011; Seifer and MacLaughlin, 2007; Somunkiran et al., 2007; Streuli et al., 2008; Van den Berg et al., 2010). Some other studies instead observe a reduction of AMH dosage during hormonal contraceptive use (Arbo et al., 2007; Kallio et al., 2013; Kristensen et al., 2012; Panidis et al., 2011).

During compliant COC use, follicular growth to a presumably ovulatory size is observed, and that is associated with loss of endocrine suppression that occurs during the HFI (Baerwald et al., 2004).

The shorter HFI in COC with low-dose preparations may increase the margin of safety in women. (Schlaff et al., 2004; Spona et al., 1996; Sullivan et al., 1999). There is no agreement whether the EE content (Van Heusden and Fauser, 1999) or the progestin formulation (Rible et al., 2009) determines the degree of residual ovarian activity at the beginning of HFI. The lower-dose rather than the higher-dose monophasic pill shows the same effectiveness of the multiphasic pill in its inhibition of follicular growth (Grimes et al., 1994).

The continuous COC dosing regimens result more effective in preventing the ovulation and they are associated with greater suppression of ovarian and endometrial function (Archer et al., 2009; Birtch et al., 2006; Kroll et al., 2015; Legro et al., 2008). Furthermore, a prospective randomized study shows that supplementing the standard 7-day HFI with EE 10 μg after 84 days of an extended COC, a reduction of FSH concentrations and number of growing follicles (Vandever et al., 2008).

The administration of EE in the HFI is controversial. Many authors show that the administration of EE during HFI more effectively suppresses the ovarian follicular activity (Killick et al., 1998; Kroll et al., 2015; Schlaff et al., 2004), but a recent randomized open-label trial documents that the 21/7 days of active low-dose COC preparation, with EE supplementation during the traditional HFI, shows a similar inhibition of ovarian follicular activity compared with the 24/4 day and the 21/7 day regimens (Seidman et al., 2015).

Low-dose COC use apparently does not increment the incidence of functional ovarian cysts (Holt et al., 2003; Lanes et al., 1992), and the occurrence of enlarged follicles is lower among women with a COC compared with those using a POP (Broome et al., 1995), although available studies are limited so far. Similarly, few studies investigate the ovarian function following missed or delayed doses of hormonal contraceptives. In most of these studies the follicular development up to pre-ovulatory diameter is commonly described in women delaying the first dose of their contraceptive cycle (Elomaa et al., 1998; Mulders et al., 2002; Petta et al., 2001), but a single study asserts that if the missed doses were restricted to only three pills, normal ovulation was not observed (Elomaa et al., 1998). Pierson shows that the ovulation rate was significantly reduced for the patch users compared with COC users. Contraceptive patch users showed follicular size and incidence of ovulation significantly lower than women taking COC in normal cycles and after predetermined dosing errors (Pierson et al., 2003). Another study shows that the effectiveness of oral contraception with imperfect compliance differs according to the progestin formulation used, but to establish the clinical relevance of this result, further studies are required (Creinin et al., 2002).

References

Alvarez-Sanchez, F., Brache, V., De Oca, V.M., Cochon, L., Faúndes, A., 2000. Prevalence of enlarged ovarian follicles among users of levonorgestrel subdermal contraceptive implants (Norplant). Am. J. Obstet. Gynecol. 182, 535–539.

Arbo, E., Vetori, D.V., Jimenez, M.F., Freitas, F.M., Lemos, N., Cunha-Filho, J.S., 2007. Serum anti-mullerian hormone levels and follicular cohort characteristics after pituitary suppression in the late luteal phase with oral contraceptive pills. Hum. Reprod. 22, 3192–3196.

Archer, D.F., Kovalevsky, G., Ballagh, S.A., Grubb, G.S., 2009. Ovarian activity and safety of a novel levonorgestrel/ethinyl estradiol continuous oral contraceptive regimen. Contraception 80, 245–253.

Baerwald, A.R., Pierson, R.A., 2004. Ovarian follicular development during the use of oral contraception: a review. J. Obstet. Gynaecol. Can. 26, 19–24.

Baerwald, A.R., Olatunbosun, O.A., Pierson, R.A., 2004. Ovarian follicular development is initiated during the hormone-free interval of oral contraceptive use. Contraception 70, 371–377.

Barnhart, K., Devoto, L., Pommer, R., Sir-Pettermann, T., Robinovic, J., Coutinho, E., 1997. Neuroendocrine mechanism of anovulation in users of contraceptive subdermal implant of nomegestrol acetate (uniplant). Fertil. Steril. 67, 250–255.

Bentzen, J.G., Forman, J.L., Pinborg, A., Lidegaard, Ø., Larsen, E.C., Friis-Hansen, L., Johannsen, T.H., Nyboe Andersen, A., 2012. Ovarian reserve parameters: a comparison between users and non-users of hormonal contraception. Reprod. Biomed. Online 25, 612–619.

Birtch, R.L., Olatunbosun, O.A., Pierson, R.A., 2006. Ovarian follicular dynamics during conventional vs. continuous oral contraceptive use. Contraception 73, 235–243.

Brache, V., Blumenthal, P.D., Alvarez, F., Dunson, T.R., Cochon, L., Faundes, A., 1999. Timing of onset of contraceptive effectiveness in Norplant implant users. II. Effect on the ovarian function in the first cycle of use. Contraception 59, 245–251.

Brache, V., Massai, R., Mishell, D.R., Moo-Young, A.J., Alvarez, F., Salvatierra, A.M., Cochon, L., Croxatto, H., Robbins, A., Faundes, A., 2000. Ovarian function during use of Nestorone(R) subdermal implants. Contraception 61, 199–204.

Broome, M., Clayton, J., Fotherby, K., 1995. Enlarged follicles in women using oral contraceptives. Contraception 52, 13–16.

Creinin, M.D., Lippman, J.S., Eder, S.E., Godwin, A.J., Olson, W., 2002. The effect of extending the pill-free interval on follicular activity: triphasic norgestimate/35 micro g ethinyl estradiol versus monophasic levonorgestrel/20 micro g ethinyl estradiol. Contraception 66, 147–152.

Crosignani, P.G., Testa, G., Vegetti, W., Parazzini, F., 1996. Ovarian activity during regular oral contraceptive use. Contraception 54, 271–273.

Deb, S., Campbell, B.K., Pincott-Allen, C., Clewes, J.S., Cumberpatch, G., Raine-Fenning, N.J., 2012. Quantifying effect of combined oral contraceptive pill on functional ovarian reserve as measured by serum anti-Müllerian hormone and small antral follicle count using three-dimensional ultrasound. Ultrasound Obstet. Gynecol. 39, 574–580.

Duijkers, I.J., Klipping, C., Verhoeven, C.H., Dieben, T.O., 2004. Ovarian function with the contraceptive vaginal ring or an oral contraceptive: a randomized study. Hum. Reprod. 19, 2668–2673.

Duijkers, I.J., Heger-Mahn, D., Drouin, D., Skouby, S., 2015. A randomised study comparing the effect on ovarian activity of a progestogen-only pill (POP) containing desogestrel and a new POP containing drospirenone in a 24/4 regimen. Eur. J. Contracept. Reprod. Health Care 20, 419–427.

Egarter, C., Putz, M., Strohmer, H., Speiser, P., Wenzl, R., Huber, J., 1995. Ovarian function during low-dose oral contraceptive use. Contraception 51, 329–333.

Elomaa, K., Rolland, R., Brosens, I., Moorrees, M., Deprest, J., Tuominen, J., Lähteenmäki, P., 1998. Omitting the first oral contraceptive pills of the cycle does not automatically lead to ovulation. Am. J. Obstet. Gynecol. 179, 41–46.

Fauser, B.C., Van Heusden, A.M., 1997. Manipulation of human ovarian function: physiological concepts and clinical consequences. Endocr. Rev. 18, 71–106.

Fitzgerald, C., Feichtinger, W., Spona, J., Elstein, M., Lüdicke, F., Müller, U., Williams, C., 1994. A comparison of the effects of two monophasic low dose oral contraceptives on the inhibition of ovulation. Adv. Contracept. 10, 5–18.

Grimes, D.A., Godwin, A.J., Rubin, A., Smith, J.A., Lacarra, M., 1994. Ovulation and follicular development associated with three low-dose oral contraceptives: a randomized controlled trial. Obstet. Gynecol. 83, 29–34.

Heger-Mahn, D., Warlimont, C., Faustmann, T., Gerlinger, C., Klipping, C., 2004. Combined ethinylestradiol/gestodene contraceptive patch: two-center, open-label study of ovulation inhibition, acceptability and safety over two cycles in female volunteers. Eur. J. Contracept. Reprod. Health Care 9, 173–181.

Holt, V.L., Daling, J.R., McKnight, B., Moore, D., Stergachis, A., Weiss, N.S., 1992. Functional ovarian cysts in relation to the use of monophasic and triphasic oral contraceptives. Obstet. Gynecol. 79, 529–533.

Holt, V.L., Cushing-Haugen, K.L., Daling, J.R., 2003. Oral contraceptives, tubal sterilization, and functional ovarian cyst risk. Obstet. Gynecol. 102, 252–258.

Hoogland, H.J., Skouby, S.O., 1993. Ultrasound evaluation of ovarian activity under oral contraceptives. Contraception 47, 583–590.

Jokubkiene, L., Sladkevicius, P., Valentin, L., 2012. Ovarian size and vascularization as assessed by three-dimensional grayscale and power Doppler ultrasound in asymptomatic women 20–39 years old using combined oral contraceptives. Contraception 86, 257–267.

Kallio, S., Puurunen, J., Ruokonen, A., Vaskivuo, T., Piltonen, T., Tapanainen, J.S., 2013. Antimullerian hormone levels decrease in women using combined contraception independently of administration route. Fertil. Steril. 99, 1305–1310.

Killick, S.R., Fitzgerald, C., Davis, A., 1998. Ovarian activity in women taking an oral contraceptive containing 20 microg ethinyl estradiol and 150 microg desogestrel: effects of low estrogen doses during the hormone-free interval. Am. J. Obstet. Gynecol. 179, 18–24.

Koering, M.J., Danforth, D.R., Hodgen, G.D., 1991. Early folliculogenesis in primate ovaries: testing the role of estrogen. Biol. Reprod. 45, 890–897.

Koering, M.J., Danforth, D.R., Hodgen, G.D., 1994. Early follicle growth in the juvenile macaca monkey ovary: the effects of estrogen priming and follicle-stimulating hormone. Biol. Reprod. 50, 686–694.

Kristensen, S.L., Ramlau-Hansen, C.H., Andersen, C.Y., Ernst, E., Olsen, S.F., Bonde, P.J., Vested, A., Toft, G., 2012. The association between circulating levels of antimullerian hormone and follicle number, androgens, and menstrual cycle characteristics in young women. Fertil. Steril. 97, 779–785.

Kroll, R., Seidman, L., Ricciotti, N., Howard, B., Weiss, H., 2015. A phase 1, multicentre, open-label study to evaluate ovarian follicular activity and hormone levels with an extended-regimen combined oral contraceptive with low-dose ethinyl estradiol supplementation. Eur. J. Contracept. Reprod. Health Care 20, 249–258.

Landgren, B.M., Diczfalusy, E., 1980. Hormonal effects of the 300 μg norethisterone (NET) minipill. Contraception 21, 87–113.

Lanes, S.F., Birmann, B., Walker, A.M., Singer, S., 1992. Oral contraceptive type and functional ovarian cysts. Am. J. Obstet. Gynecol. 166, 956–961.

Legro, R.S., Pauli, J.G., Kunselman, A.R., Meadows, J.W., Kesner, J.S., Zaino, R.J., Demers, L.M., Gnatuk, C.L., Dodson, W.C., 2008. Effects of continuous versus cyclical oral contraception: a randomized controlled trial. J. Clin. Endocrinol. Metab. 93, 420–429.

Li, H.W.R., Wongb, C.Y.G., Yeunga, W.S.B., Ho, P.C., Ng, E.H., 2011. Serum anti-müllerian hormone level is not altered in women using hormonal contraceptives. Contraception 83, 582–585.

Mäkäräinen, L., van Beek, A., Tuomivaara, L., Asplund, B., Coelingh Bennink, H., 1998. Ovarian function during the use of a single contraceptive implant: implanon compared with Norplant. Fertil. Steril. 69, 714–721.

McCann, M.F., Potter, L.S., 1994. Progestin-only oral contraception: a comprehensive review. Contraception 50 (Suppl. 1), S9–S195.

Mulders, T.M., Dieben, T.O., Bennink, H.J., 2002. Ovarian function with a novel combined contraceptive vaginal ring. Hum. Reprod. 17, 2594–2599.

Panidis, D., Georgopoulos, N.A., Piouka, A., Katsikis, I., Saltamavros, A.D., Decavalas, G., Kandarakis, E.D., 2011. The impact of oral contraceptives and metformin on anti-Müllerian hormone serum levels in women with polycystic ovary syndrome and biochemical hyperandrogenemia. Gynecol. Endocrinol. 27, 587–592.

Petta, C.A., Faúndes, A., Dunson, T.R., Ramos, M., DeLucio, M., Faúndes, D., Bahamondes, L., 1998. Timing of onset of contraceptive effectiveness in Depo-Provera users. II. Effects on ovarian function. Fertil. Steril. 70, 817–820.

Petta, C.A., Hays, M., Brache, V., Massai, R., Hua, Y., Alvarez-Sánchez, F., Croxatto, H., d'Arcangues, C., Cook, L.A., Bahamondes, L., 2001. Delayed first injection of the once-a-month injectable contraceptive containing 25 mg of medroxyprogesterone acetate and 5 mg of E(2)-cypionate: effects on ovarian function. Fertil. Steril. 75, 744–748.

Pierson, R.A., Archer, D.F., Moreau, M., Shangold, G.A., Fisher, A.C., Creasy, G.W., 2003. Ortho Evra/Evra versus oral contraceptives: follicular development and ovulation in normal cycles and after an intentional dosing error. Fertil. Steril. 80, 34–42.

Rabe, T., Nitsche, D.C., Runnebaum, B., 1997. The effects of monophasic and triphasic oral contraceptives on ovarian function and endometrial thickness. Eur. J. Contracept. Reprod. Health Care 2, 39–51.

Rible, R.D., Taylor, D., Wilson, M.L., Stanczyk, F.Z., Mishell, D.R., Jr., 2009. Follicular development in a 7-day versus 4-day hormone-free interval with an oral contraceptive containing 20 mcg ethinyl estradiol and 1 mg norethindrone acetate. Contraception 79, 182–188.

Rossmanith, W.G., Steffens, D., Schramm, G., 1997. A comparative randomized trial on the impact of two low-dose oral contraceptives on ovarian activity, cervical permeability, and endometrial receptivity. Contraception 56, 23–30.

Schlaff, W.D., Lynch, A.M., Hughes, H.D., Cedars, M.I., Smith, D.L., 2004. Manipulation of the pill-free interval in oral contraceptive pill users: the effect on follicular suppression. Am. J. Obstet. Gynecol. 190, 943–951.

Seidman, L., Kroll, R., Howard, B., Ricciotti, N., Hsieh, J., Weiss, H., 2015. Ovulatory effects of three oral contraceptive regimens: a randomized, open-label, descriptive trial. Contraception 91, 495–502.

Seifer, D.B., MacLaughlin, D.T., 2007. Mullerian Inhibiting Substance is an ovarian growth factor of emerging clinical significance. Fertil. Steril. 88, 539–546.

Shaaban, M.M., Segal, S., Salem, H.T., Ghaneimah, S.A., Khalifa, E.A., Ahmed, A.G., 1993. Sonographic assessment of ovarian and endometrial changes during long-term Norplant use and their correlation with hormonal levels. Fertil. Steril. 59, 998–1002.

Somunkiran, A., Yavuz, T., Yucel, O., Ozdemir, I., 2007. Antimullerian hormone levels during hormonal contraception in women with polycystic ovary syndrome. Eur. J. Obstet. Gynecol. Reprod. Biol. 134, 196–201.

Spellacy, W.N., Kalra, P.S., Buhi, W.C., Birk, S.A., 1980. Pituitary and ovarian responsiveness to a graded gonadotropin releasing factor stimulation test in women using a low-estrogen or a regular type of oral contraceptive. Am. J. Obstet. Gynecol. 137, 109–115.

Spona, J., Elstein, M., Feichtinger, W., Sullivan, H., Lüdicke, F., Müller, U., Düsterberg, B., 1996. Shorter pill-free interval in combined oral contraceptives decreases follicular development. Contraception 54, 71–77.

Spona, J., Binder, N., Höschen, K., Feichtinger, W., 2010. Suppression of ovarian function by a combined oral contraceptive containing 0.02 mg ethinyl estradiol and 2 mg chlormadinone acetate given in a 24/4-day intake regimen over three cycles. Fertil. Steril. 94, 1195–1201.

Streuli, I., Fraisse, T., Pillet, C., Ibecheole, V., Bischof, P., de Ziegler, D., 2008. Serum antimullerian hormone levels remain stable throughout the menstrual cycle and after oral or vaginal administration of synthetic sex steroids. Fertil. Steril. 90, 395–400.

Sullivan, H., Furniss, H., Spona, J., Elstein, M., 1999. Effect of 21-day and 24-day oral contraceptive regimens containing gestodene

(60 microg) and ethinyl estradiol (15 microg) on ovarian activity. Fertil. Steril. 72, 115–120.
Tafurt, C.A., Sobrevilla, L.A., de Estrada, R., 1980. Effects of progestin-estrogen combination and progestational contraceptives on pituitary gonadotropins, gonadal steroids and sex hormone-binding globulin. Fertil. Steril. 33, 261–266.
Tayob, Y., Adams, J., Jacobs, H.S., Guillebaud, J., 1985. Ultrasound demonstration of increased frequency of functional ovarian cysts in women using progestogen-only oral contraception. Br. J. Obstet. Gynaecol. 92, 1003–1009.
Teichmann, A.T., Brill, K., Albring, M., Schnitker, J., Wojtynek, P., Kustra, E., 1995. The influence of the dose of ethinylestradiol in oral contraceptives on follicle growth. Gynecol. Endocrinol. 9, 299–305.
Van den Berg, M.H., Van Dulmen-den Broeder, E., Overbeek, A., Twisk, J.W.R., Schats, R., Van Leeuwen, F.E., Kaspers, G.J., Lambalk, C.B., 2010. Comparison of ovarian function markers in users of hormonal contraceptives during the hormone free interval and subsequent natural early follicular phases. Hum. Reprod. 25, 1520–1527.
Van der Does, J., Exalto, N., Dieben, T., Bennink, H.C., 1995. Ovarian activity suppression by two different low-dose triphasic oral contraceptives. Contraception 52, 357–361.
Van Heusden, A.M., Fauser, B.C., 1999. Activity of the pituitary-ovarian axis in the pill-free interval during use of low-dose combined oral contraceptives. Contraception 59, 237–243.
Vandever, M.A., Kuehl, T.J., Sulak, P.J., Witt, I., Coffee, A., Wincek, T.J., Reape, K.Z., 2008. Evaluation of pituitary-ovarian axis suppression with three oral contraceptive regimens. Contraception 77, 162–170.
Wan, L.S., Ganguly, M., Weiss, G., 1981. Pituitary response to LHRH stimulation in women on oral contraceptives: a follow up dose response study. Contraception 24, 229–234.
Young, R.L., Snabes, M.C., Frank, M.L., Reilly, M., 1992. A randomized, double-blind, placebo-controlled comparison of the impact of low-dose and triphasic oral contraceptives on follicular development. Am. J. Obstet. Gynecol. 167, 678–682.

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