Immediate pre-ovulatory administration of 30 mg ulipristal acetate significantly delays follicular rupture

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BACKGROUND: Current methods of hormonal emergency contraception (EC) are ineffective in preventing follicular rupture when administered in the advanced pre-ovulatory phase. This study was designed to determine the capacity of ulipristal acetate (UPA), a selective progesterone receptor modulator developed for EC, to block follicular rupture when administered with a follicle of \( \geq 18 \) mm.

METHODS: This was a double-blind, crossover, randomized, placebo-controlled study. Thirty-five women contributed with UPA (30 mg, oral) and a placebo cycle. Serial blood sampling for luteinizing hormone (LH), estradiol and progesterone measurements and follicular monitoring by ultrasound were performed before and for 5 days following treatment. Follicular rupture inhibition was assessed in the overall study population and in subgroups of women stratified by when treatment was administered in relation to LH levels (before the onset of the LH surge, after the onset of the surge but before the LH peak or after the LH peak).

RESULTS: Follicular rupture failed to occur for at least 5 days following UPA administration in 20/34 cycles [59%; 95% confidence interval (CI) (40.7–75.4%)], whereas rupture took place in all cycles within 5 days of placebo intake. When UPA was administered before the onset of the LH surge, or after the onset but before the LH peak, follicle rupture had not occurred within 5 days in 8/8 (100%) and 11/14 [78.6%; 95% CI (49.2–95.3%)] cycles, respectively. In contrast, when UPA was given after the LH peak, follicle rupture inhibition was only observed in 1/12 [8.3%; 95% CI (0.2–38.5%)] cycles.

CONCLUSIONS: This study demonstrates that UPA can significantly delay follicular rupture when given immediately before ovulation. This new generation EC compound could possibly prevent pregnancy when administered in the advanced follicular phase, even if LH levels have already begun to rise, a time when levonorgestrel EC is no longer effective in inhibiting ovulation.

NCT01107093: Comparison of CDB-2914 versus placebo in the prevention of follicular rupture post-LH surge.

Key words: ulipristal acetate / emergency contraception / follicular rupture / LH surge

Introduction

The most widely used emergency contraception (EC) regimen is the progestogen levonorgestrel (LNG) at the dose of 1.5 mg taken within 72 h after unprotected intercourse. Within this time frame, LNG-EC is reasonably effective in preventing unwanted pregnancy with 1.5–2.6% pregnancy rates after a single dose when taken between 0–24 h and 48–72 h after unprotected intercourse, respectively (Von Hertzen et al., 1998; Piaggio et al., 1999; Cheng et al., 2008).

Since the first Yuzpe EC regimen described more than 30 years ago (Yuzpe et al., 1974), understanding of the mechanism of action of hormonal EC has gradually grown but remains limited. The LNG-EC regimen acts to prevent pregnancy by blocking or delaying the luteinizing hormone (LH) surge. The more advanced in the follicular phase that LNG is taken, the lower its ability to block ovulation and thus prevent fertilization after unprotected intercourse (Durand et al., 2001; Hapangama et al., 2001; Marions et al., 2002; Croxatto et al., 2004; Massai et al., 2007; Novikova et al., 2007). Thus, the ability of LNG to interfere with the ovulatory process is limited to its...
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administration during the period preceding the onset of the LH surge. Once the ovulatory process has been triggered by the LH surge, this progestogen agent cannot prevent the follicle from rupturing and releasing the oocyte, an event that normally takes place 36 h later.

Ulipristal acetate (UPA) (formerly known as CDB-2914), a selective progesterone receptor modulator specifically developed for EC, was at least as effective as LNG in prevention of pregnancy when administered up to 3 days (72 h) after unprotected intercourse (Creinin et al., 2006). In a large recent randomized single-blind non-inferiority multicentre trial comparing a single dose of 30 mg UPA with 1.5 LNG, efficacy was evaluated in 1899 women. Pregnancy rates were 1.8% [95% confidence interval (CI) 1.0–3.0] in the UPA group and 2.6% (95% CI 1.7–3.9) in the LNG groups when treatment was administered within 72 h of intercourse in 1696 women. Comparing the efficacy of UPA and LNG using a meta-analysis of the two above studies, the odds ratio (OR) of pregnancy for UPA versus LNG when taken within 0–120 h after intercourse was 0.55 (95% CI 0.32–0.93). This superiority was significant at 24, 72 and 120 h between intercourse and treatment (Glasier et al., 2010). In another study, a pregnancy rate of 2.1% (95% CI 1.4–3.1) was reported among 1241 women who received UPA (30 mg) when presenting for EC 48–120 h after unprotected intercourse (Fine et al., 2010).

The mechanism by which UPA interferes with ovulation has not yet been elucidated: it could act through inhibition or delay of the LH surge, or act directly on the ovary to inhibit follicular rupture. Stratton et al. (2000) showed that a single mid-follicular dose of 10–100 mg of UPA (CDB-2914) administered with a follicle of 14–16 mm, caused a dose-dependent delay in the time interval from treatment to follicular rupture and suppression of estradiol (E2). In a recent study using the ovarian gene expression profiling in mice subjected to gonadotrophin-induced superovulation, the administration of UPA 1 h before hCG administration resulted in a >90% inhibition of oocyte release, strongly suggesting a direct effect of the compound upon dominant follicles (Palanisamy et al., 2006). Delaying follicular rupture, which normally occurs on average 36 h after the onset of the LH surge, could be a possible mechanism of action of UPA that could provide a wider time window of action and a higher efficacy at peak fertility time than LNG.

The present study was designed to characterize the capacity of a single oral 30 mg dose of UPA (corresponding to the product recently marketed for EC in Europe) to inhibit follicular rupture when given at a very late pre-ovulatory stage (when the size of the lead follicle had reached 18 mm) in comparison to placebo administered in the same women. The study also investigated the efficacy of UPA in preventing follicular rupture when given before or after onset of the LH surge.

**Materials and Methods**

The study was conducted in two large reproductive health clinics in Latin America (ICMER in Santiago, Chile, and PROFAMILIA in Santo Domingo, Dominican Republic) between May and December 2008. Approval was granted for the study protocol and subject information and consent forms by the Ethics Committee of each centre.

A total of 46 women in good general health were screened for inclusion in the study after they gave written informed consent. To be eligible they had to be 18–35 years old, with regular menstrual cycles in the past 3 months, non-pregnant and non-breastfeeding, not currently using hormonal contraception, and protected from pregnancy by tubal ligation or non-hormonal intrauterine device (IUD).

**Study design**

This was a double-blind, crossover, randomized, placebo-controlled study designed to evaluate the effect of a single oral dose of 30 mg UPA on the outcome of the leading ovarian follicle when administered immediately before ovulation, i.e. when the follicle was ≥18 mm in diameter. Following a screening visit, eligible women were followed for four complete menstrual cycles: a first treatment cycle (Cycle 1) followed by two wash-out cycles (Cycles 2 and 3) and a second treatment cycle (Cycle 4). Women were randomized to receive UPA or placebo in a crossover fashion in the treatment cycles. Randomization schedule was generated by an independent investigational medical products supplier (Creapharm, France) and stored in a sealed envelope with HRA Pharma’s qualified person until the database quality was entirely verified (inconsistent and missing data were queried to study sites) and locked (after database lock on 10 April 2009, no further data change were made by either sites, sponsor or data management staff). If screened women satisfied all inclusion/exclusion criteria, they entered the study and underwent ultrasound monitoring starting from Day 5 to 8 of their next menstrual cycle (Cycle 1) until such time as the lead follicle reached ≥18 mm, at which time they were randomized and treatment was given in front of study staff. From that time on, they were monitored daily by ultrasound and hormone assays until the fifth day following treatment, then twice a week until menses. Following two washout cycles during which women received no treatment, the same procedure was repeated in Cycle 4. An end-of-study visit was performed at the beginning of the menses that followed Cycle 4 and included a trans-vaginal ultrasound (TVU). If the last ultrasound examination showed a follicle or cyst larger than 25 mm in diameter, a TVU was repeated once a week after menses until resolution.

**Study medication**

The study medication was a single oral tablet of either UPA (30 mg) or matching placebo administered in a double-blind, crossover fashion. The placebo treatment cycle served as a reference to the UPA treatment cycle for each of the 35 subjects enrolled. UPA and placebo pills were obtained from HRA Pharma (Paris, France) and packaged into identical blisters and boxes numbered to match an ad hoc randomization list.

**Ultrasonography**

To assess the presence of follicular rupture, a TVU was performed both in menstrual Cycle 1 (first treatment cycle) and in menstrual Cycle 4 (second treatment cycle) three times a week starting at Day 5–8 until the leading follicle was ≥15 mm, then daily until the fifth day after treatment administration, and thereafter twice a week until menses. Follicular rupture was defined as an abrupt disappearance (or >50% reduction in size) of the leading follicle whose mean diameter was 15–25 mm in the TVU performed on the day before. When follicular rupture occurred after the 5-day period of daily visits, the day of follicular rupture was assumed conservatively to occur on the first day following the last observation of the dominant follicle. For example, if a dominant follicle was observed on Day 5 following treatment and had ruptured on the following observation occurring on Day 9, follicular rupture was recorded as occurring on Day 6.

At the Chilean clinic, a TVU was performed by two ultrasonographers specifically trained in obstetrics and gynaecology ultrasound using a Medison SA 6000C or ALOKA SSD-3500SX ultrasound scanner system, with a 7.5-MHz vaginal transducer (Sony Corp., Tokyo, Japan). At the Dominican clinic, all TVUs were performed by a single highly experienced ultrasonographer using a Shimadzu SDU-400 with a 4–8 MHz vaginal transducer.
Hormonal assays
Levels of LH, E2 and progesterone were measured daily starting when the leading follicle was ≥15 mm until the fifth day after treatment administration, during both treatment cycles. Additionally, progesterone levels were measured twice a week until menses during the two treatment cycles, and once 6–8 days before the expected menses during both washout cycles.

LH, E2 and progesterone were assayed centrally in Santiago, Chile, using standardized laboratory procedures. Serum LH was measured using enzyme immunoassay (EIA, Immunometric, UK Ltd.). For low-, medium- and high-quality control samples, the inter-assay coefficient of variation was 6.0, 5.6 and 7.9%, respectively, and the intra-assay coefficient of variation was 2.6 and 2.3%, for low- and high-quality control samples, respectively. E2 and progesterone were measured using a radio-immunoassay (DPC, Diagnostic Products Corporation, Los Angeles, CA, USA). All samples from the same subject were assayed in the same run.

Data analysis
The primary end-point was follicular rupture inhibition, defined as persistence of the unruptured dominant follicle. Secondary end-points included: LH (UI/L), E2 (pmol/L) and progesterone (nmol/L) levels. The LH surge onset and LH peak definitions were calculated based on LH levels measured in 100 ovulatory placebo cycles performed in the same two investigation sites during previous studies with similar design. The presence of an LH surge onset was defined as an LH increase by at least 40% compared with the day before and greater than 6 IU/L, OR over 8 IU/L for the first time; while LH peak was defined as an LH value >15.6 IU/L. Progesterone level after treatment intake was defined as luteal if at least two consecutive levels were >10 nmol/L, OR as anovulatory if levels were constantly under 10 nmol/L. Another secondary end-point was the mean time elapsed from the treatment intake day to observed rupture of the leading follicle.

A minimum sample size of 30 completed subjects was estimated in order to reach at least 80% power for the primary efficacy analysis, based on follicular rupture inhibition results from four previous similar studies in which less than 15% of women given a placebo on the day their leading follicle reached 18 mm failed to have follicular rupture within 5 days as compared with 0–50% in women receiving EC (Croxatto et al., 2002, 2004; Massai et al., 2007; Brache et al., 2009).

The primary statistical analysis tested the null hypothesis that the proportion of subjects with an inhibition of follicular rupture on a given day after treatment administration is equal between treatment groups with a significance level of 5% (two-sided). The statistical test performed was a McNemar’s test for a crossover design using paired dichotomous variable ‘inhibition of follicle rupture’. For secondary continuous variables such as cycle length and hormonal levels (LH, progesterone and E2), the treatment effect was tested in an analysis of covariance model including cofactors of treatment, cycle (Cycle 1 versus Cycle 4), sequence (placebo/UPA versus UPA/placebo) and subject nested within sequence. Effects were adjusted to baseline values as a covariate factor. Frequency tables for discrete variables were analysed by MH-χ² statistics. Time from treatment intake to follicular rupture was analysed in a survival analysis. All statistical analyses were performed with the SAS® system version 9.1.

Results
Baseline characteristics
Out of the 46 screened women, 35 were enrolled, randomized and completed the study (22 in Chile and 13 in Dominican Republic).

Ten screened subjects did not fulfill the eligibility criteria and one subject was not randomized because ovulation occurred before the follicle reached the size of 18 mm. One subject had an unclear follicular status at the end of both treatment cycles and was excluded, reducing to 34 efficacy evaluable subjects (Fig. 1).

The baseline demographic and gynaecological history characteristics for both sites were similar for most variables. The mean age was 31.0 ± 3.5 years (range 22–35), all women had normal menstrual cycles with a mean of 28.8 ± 1.8 days duration (range 24–28) and all had had previous pregnancies. BMI was significantly different in the two sites, 24.4 ± 2.9 kg/m² in Chile and 27.5 ± 3.6 kg/m² in the Dominican Republic (P = 0.0075). The current contraceptive method was tubal lig-ation for 100% of the Dominican Republic and 72.7% of Chile subjects, while the remaining subjects in Chile used IUDs.

All enrolled women had a normal TVU at baseline. There were no differences between mean follicular diameter (18.4 ± 0.6 versus 18.5 ± 0.8 mm), mean E2 levels (536.8 ± 182.8 versus 519.5 ± 181.5 pmol/L) and mean progesterone levels (2.4 ± 1.5 versus 2.7 ± 1.5 nmol/L) at the time of UPA and placebo administration, respectively. Cycle day of treatment was also similar (13.8 ± 12.6 versus 13.5 ± 2.9) for UPA and placebo. LH status at the time of treatment administration was similar in both placebo and UPA cycles (Table I). Twenty-six subjects were treated with UPA after the LH onset or peak (regardless of the treatment timing in relation to the LH in the placebo cycle). Of these, 19 subjects received both treatment and placebo after the onset of the LH surge or peak.

Follicular rupture during the post-treatment period
In placebo cycles, all dominant follicles had ruptured by 5 days after treatment. In contrast, the dominant follicle persisted for at least 5 days in 20/34 (58.8%) UPA cycles. The difference between UPA and placebo was highly significant (P < 0.0001) (Table II). The magnitude of inhibition of follicular rupture differed according to the LH status at the time of treatment. When UPA was administered before the LH surge onset, the dominant follicle was still present in Figure 1 Flow chart for study of the capacity of UPA to block follicular rupture. mITT (modified intention to treat) population included women that were enrolled, randomized, received both treatments, completed Cycle 4 and had a known follicular status after treatment intake in both cycles. The mITT data set is the primary efficacy population for statistical analysis.
Table I LH status and mean LH levels (UI/l) in women at time of treatment with UPA or placebo in a study of the capacity of UPA to block follicular rupture.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>UPA</th>
<th>Placebo</th>
</tr>
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<tbody>
<tr>
<td>n (1st/2nd cycle)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>LH (Mean ± SD)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Treatment before LH surge onset</td>
<td>8 (5/3)</td>
<td>4.1 ± 1.8</td>
</tr>
<tr>
<td>Treatment after LH surge onset but before LH peak</td>
<td>14 (7/7)</td>
<td>10.0 ± 2.6</td>
</tr>
<tr>
<td>Treatment after LH peak</td>
<td>12 (5/7)</td>
<td>54.8 ± 21.3</td>
</tr>
</tbody>
</table>

Table II Inhibition of follicular rupture observed 5 days after treatment administration.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>UPA n (%)</th>
<th>Placebo n (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Placebo n (%)</td>
<td>No (0%)</td>
<td>Yes (14 (41.2%))</td>
</tr>
<tr>
<td>No</td>
<td>14 (41.2%)</td>
<td>20 (58.8%)</td>
</tr>
<tr>
<td>Yes</td>
<td>0 (0%)</td>
<td>0 (0%)</td>
</tr>
<tr>
<td>McNemar’s test</td>
<td>Statistic (DF)</td>
<td>20.0000 (1)</td>
</tr>
<tr>
<td>P-value</td>
<td>&lt;0.0001</td>
<td></td>
</tr>
</tbody>
</table>

Table III Inhibition of follicular rupture at 5 days after treatment administration, stratified by LH status at time of treatment.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>UPA n (%) [95% CI]</th>
<th>Placebo n (%) [95% CI]</th>
</tr>
</thead>
<tbody>
<tr>
<td>Treatment before LH surge onset</td>
<td>8/8 (100%)</td>
<td>0/12 (0%)</td>
</tr>
<tr>
<td>Treatment after LH surge onset but before LH peak</td>
<td>11/14 (78.6%) [49.2–95.3%]</td>
<td>0/6 (0%)</td>
</tr>
<tr>
<td>Treatment after LH peak</td>
<td>1/12 (8.3%) [0.2–38.5]</td>
<td>0/16 (0%)</td>
</tr>
</tbody>
</table>

CI, confidence interval.

100% (8/8) of cycles at 5 days. When given after the LH surge but prior to the peak, follicular rupture inhibition in the 5-day period was 78.6% (11/14). In contrast, when UPA was given after the LH peak level was reached, the follicle rupture inhibition was observed only in 1/12 (8.3%) of UPA cycles (Table III).

Time (in days) from treatment intake to follicular rupture has been analysed in a survival analysis and survival probabilities are displayed in Fig. 2. Follicular rupture occurred significantly later following UPA treatment (6 days in median) compared with placebo (2 days in median) (P = 0.0279). Following placebo administration, follicular rupture occurred within 72 h of treatment intake in 26/34 (76.5%) cycles, with only two subjects still having an unruptured follicle on Day 4 post-treatment. In contrast, follicle rupture occurred in only 11 subjects (32.3%) within 72 h after UPA treatment. Of note, in all of these cycles, the treatment had been administered on the day of LH peak. Follicular rupture was delayed by 5–10 days or did not occur in the remaining 67.6% (23/34) of cycles.

When treatment was given before the LH peak, the mean time that elapsed from treatment intake to follicle rupture was still significantly longer in UPA cycles (6.85 ± 1.42 days) than in placebo cycles (3.53 ± 0.80 days) (P = 0.0001), however, when UPA was given at the time of the LH peak, the time elapsed to rupture was similar to placebo (1.54 ± 0.52 versus 1.31 ± 0.48).

Follicular growth and outcome

Table IV shows the final outcome of the follicle at the end of the menstrual cycle. Of the 20 UPA cycles in which follicular rupture had not occurred by Day 5, 15 women had a delayed rupture (Day 6–10) and the mean follicular diameter prior to rupture was larger (24.3 ± 2.8 mm) than the one observed in the placebo group (20.5 ± 2.5 mm) or in the UPA cycles (19.8 ± 2.5 mm) that ruptured within 5 days of treatment. Two women had abnormal cycles with luteinization occurring prior to delayed rupture on Day 10 post-UPA (progesterone = 12.8 and 60.7 nmol/l on the sample pre-rupture), and in three women a luteinized unruptured follicle was documented. These follicles measured a maximum of 52.5, 30.0 and 22.0 mm, but all had decreased to < 25 mm diameter by the onset of menses.

Hormones

An immediate drop in LH levels was observed on the day following UPA intake, from a mean of 25.3 ± 28.1 to 5.7 ± 7.9 IU/l, whereas no such change was observed following placebo intake (26.3 ± 26.2 to 23.6 ± 25.4). Figure 3 shows mean LH, E2 and progesterone levels after UPA and placebo treatment, categorized by the LH status at the time of treatment. Excluding the cycles in which an LH peak was already present on the day of treatment, an LH peak level was reached in all placebo cycles by Day 3 after treatment administration, while no LH peak was seen in the UPA cycles until 4–7 days after UPA intake.

Mean E2 levels during placebo cycles were 370.4 ± 207.5 and 420.4 ± 246.6 pmol/l across UPA cycles (P = 0.002), this effect was independent of treatment cycle sequence. This difference is very likely explained by the longer lifespan of the growing dominant follicle actively producing E2 during UPA cycles. E2 levels initially drop for the first 3 days but then increase again, grossly following follicular growth (Fig. 3). In the cycles in which UPA or placebo were administered with a pre-existing LH peak, a significant pre-ovulatory progesterone increase was present on the day of treatment (4.0 ± 1.4 and 3.8 ± 1.3 nmol/l, respectively), in contrast with a mean progesterone level of 1.6 ± 0.6.
and 1.7 ± 0.7 nmol/l, when UPA or placebo were administered before the LH peak. Progesterone levels >10 nmol/l were observed in all UPA and placebo cycles, however, progesterone rise was significantly delayed after the UPA treatment following the delayed follicular rupture (Fig. 3). The mean highest progesterone level during UPA cycles was 49.2 ± 18.3 nmol/l similar to 53.5 ± 16.0 nmol/l for placebo cycles.

**Menstrual cycle length**

Menstrual cycle length (adjusted for baseline cycle duration) was significantly increased after UPA treatment compared with placebo (32.7 ± 3.7 and 30.2 ± 4.1 days, respectively, P = 0.0024).

**Safety profile**

The adverse events reported during this study were few and similar to those described in the recently published Phases 2 and 3 studies (Creinin et al., 2006; Glasier et al., 2010). Furthermore, adverse events were reported more frequently in the placebo than UPA cycles. There was no serious adverse event reported during the trial, and no discontinuation because of an adverse event.

**Discussion**

The results of the present study demonstrate that a single dose of 30 mg UPA administered immediately before ovulation (either before the LH surge has begun or when the LH surge has already begun) significantly delays or inhibits subsequent follicular rupture in comparison to placebo-treated cycles. An intact follicle is still present on the fifth day after UPA administration in almost 60% of women and in none of the placebo.

Ovarian hormones mirrored the pattern of follicular development observed. E2 levels during UPA cycles were significantly higher, likely a result of the prolonged follicular activity. Progesterone levels were within the normal luteal phase range in all but two of the UPA cycles; when follicular rupture was delayed, progesterone rise was also delayed in comparison with the placebo cycle, but remained within a normal range.

The effect of treatment on menstrual cycle length (increased by 2.5 days in UPA cycles compared with baseline and placebo cycles) was very similar to results from previous efficacy studies (Creinin et al., 2006; Fine et al., 2010; Glasier et al., 2010). This increase in cycle length can be explained mostly by the delay in follicular rupture observed in this study after UPA intake.

It is important to note that although follicular rupture is delayed by 4–10 days after UPA intake, ovarian hormone production resumes and follicular rupture indeed takes place in most of the cycles, but later than expected. Although the effect of the delay in follicle development and rupture on oocyte maturation is unknown, further unprotected intercourse after UPA for EC may be potentially fertile, resulting in an increase in pregnancy risk.
The design of this study allowed evaluation of the pharmacodynamic effect of UPA under controlled conditions, mainly (i) the treatment was always administered on the day the size of the leading follicle reached 18 mm, (ii) UPA and placebo treatments were compared within the same subject (crossover design) and (iii) the timing of the treatment intake in relation to the LH surge onset was documented.

Figure 3 LH, estradiol and progesterone levels across treatment cycle stratified by LH status at the time of treatment administration.
This approach ensured that the treatment was the only major intervention. In previous studies evaluating LNG-EC, these methods were able to detect significant differences between the tested product and placebo (Croxatto et al., 2004; Massai et al., 2007).

Because initiation of the ovulatory process is not perfectly correlated to follicular diameter, this study allowed evaluation of the effect of UPA treatment in different pre-ovulatory stages in spite of the fact that all treatments were administered with a follicle \( \geq 18 \) mm. In some women, LH levels had not begun to rise at the time of UPA treatment, in others the LH surge had begun but not reached peak levels, and in others the LH peak had been reached. Our results show that the ability of UPA to interfere with follicular rupture appears to depend on when the drug is administered in relation to LH levels. When administered before the onset of the LH surge, UPA delayed the LH peak and follicular rupture in all cycles; when administered after onset of the LH surge but before the LH peak, the magnitude of the effect was still significant. In contrast, in the cycles in which the UPA treatment took place on the day of the LH peak, when a significant rise in peak had already occurred, follicle rupture followed within 24–48 h with the exception of one woman who exhibited a luteinized unruptured follicle.

The present results indicate that follicular rupture delay is mainly mediated by postponement of LH peak. Nonetheless, UPA may also have a direct effect on the dominant follicle by interfering with progesterone receptor regulated pathways that modulate ovulation, as has been demonstrated in mice (Palanisamy et al., 2006).

Stratton et al. (2000) showed that a single mid-follicular dose of 10–100 mg of UPA (CDB-2914) administered with a follicle of 14–16 mm, caused a dose-dependent delay in the time interval from treatment to follicular rupture and suppression of E2. At higher doses, the initial lead follicle often stopped growing and was replaced by a new lead follicle. This phenomenon was not observed in our study, and it may be related to the fact that we administered UPA later in the cycle, with a pre-ovulatory follicle \( \geq 18 \) mm, instead of 14–16 mm; another possible difference may be the lower dose used in our study.

Existing hormonal emergency contraceptives based on LNG or estrogend-progestogen combinations administered well before the onset of the LH surge exert inhibitory effects on ovulation via shunting of the LH surge, but they do not significantly delay or inhibit follicular rupture when administered in the advanced pre-ovulatory phase (Croxatto et al., 2001; Gemzell-Danielsson and Marions, 2004; Novikova et al., 2007). In two studies, conducted by the same investigators, using the same design and same conditions of treatment (administration when lead follicle has reached 18 mm in diameter), LNG-EC inhibited dominant follicular rupture for 5 days after treatment in only 2/17 (12%) and 5/31 (16%) women, respectively (Croxatto et al., 2004; Massai et al., 2007). The results from these two trials were very similar and, when combined, this resulted in follicle rupture inhibition in 7/48 women (14.6%) of the LNG studied cycles as compared with 20/34 (58.8%) women with UPA. When comparing the proportions of follicular rupture inhibition at 5 days of treatment using a Fisher exact test, the difference between LNG and UPA is significant (\( P < 0.0001 \)).

In summary, this study provides mechanistic evidence to explain how UPA could be more effective in preventing pregnancy than current reference EC methods. It suggests that UPA is able to inhibit or significantly delay follicular rupture for over 5 days if given immediately before ovulation by postponing the LH peak.

With respect to the proposed therapeutic use of UPA for EC, these results are doubly pertinent: the 5-day window is important in that it corresponds to the estimated lifespan of sperm in the female genital tract (Wilcox et al., 1995), and the immediate pre-ovulatory treatment window is relevant because intercourse at this time of the cycle carries a high probability of conception (Trussell et al., 1998; Wilcox et al., 2004). If UPA administered after mid-cycle unprotected intercourse can prevent ovulation from occurring for the following 5 days, it may result in an increased effectiveness in pregnancy prevention during a wider time window after unprotected intercourse. This new generation emergency contraceptive compound could possibly prevent pregnancy when administered very late in the follicular phase, even if the LH levels have already begun to rise, a time when LNG emergency contraceptives are no longer effective.

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