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Oral versus intramuscular progesterone for in vitro fertilization: a prospective randomized study

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Objective: To evaluate the efficacy of oral micronized progesterone compared with IM progesterone in oil for luteal support in patients undergoing IVF who are treated with a GnRH agonist.

Design: Randomized prospective clinical trial.

Setting: University-based IVF center.

Patient(s): Women <40 years of age who were undergoing IVF with luteal GnRH pituitary down-regulation.

Intervention(s): Patients were randomized to receive either oral micronized progesterone (200 mg three times daily) or IM progesterone (50 mg daily).

Main Outcome Measure(s): Progesterone levels at standardized days 21 and 28, and pregnancy and embryo implantation rates.

Result(s): Day 21 progesterone levels were 77.6 ± 13.2 ng/mL in the IM group and 81.5 ± 16.2 ng/mL in the oral group. Day 28 progesterone levels were 76.3 ± 15.0 ng/mL in the IM group and 53.6 ± 10.1 ng/mL in the oral group. The clinical pregnancy rates were 57.9% and 45.8% for the IM and oral groups, respectively. The implantation rate per embryo was significantly higher in the IM group (40.9%) than in the oral group (18.1%).

Conclusion(s): When used according to our protocols, oral progesterone and IM progesterone result in comparable levels of circulating progesterone. However, oral progesterone results in a reduced implantation rate per embryo. (Fertil Steril® 1999;71:614–8. ©1999 by American Society for Reproductive Medicine.)

Key Words: Oral micronized progesterone, in vitro fertilization, implantation

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Progesterone supplementation of the luteal phase is prescribed routinely for women undergoing IVF. The most common routes of administration are IM injection and vaginal suppository. Progesterone delivered by IM injection can lead to marked inflammation at the injection site, resulting in redness, pain, and even sterile abscess formation. Although vaginal suppositories are easier to tolerate, the suppository material may escape from the vagina, leading to inconvenience and uncertainty as to the dosage of progesterone absorbed. Progesterone taken orally would avoid these potential complications. This prospective randomized study examined the use of oral micronized progesterone for luteal support after IVF and compared its efficacy to that of IM progesterone.

MATERIALS AND METHODS

Patients were recruited through signs placed in the waiting areas at our clinic that explained the study. The inclusion criteria were the use of GnRH down-regulation and age <40 years. Patients were assigned to receive either IM or oral progesterone supplementation according to a randomization table. The protocol was approved by the institutional board of research, and all patients gave informed consent before entering the study.

Patients were prescribed either progesterone in oil (50 mg IM daily) or micronized progesterone (200 mg orally three times daily) beginning on day 15 of an IVF cycle. The day of oocyte retrieval was normalized to day 14, and

serum samples for progesterone were drawn in the morning on days 21 and 28, before the administration of any medications on those days. All patients underwent ovarian stimulation using luteal phase GnRH down-regulation followed by stimulation with IM FSH, hMG, or a combination of FSH and hMG. Embryo transfers were performed on day 3 after oocyte retrieval. Embryos were graded on a scale of 1–4, with 1 being the highest quality. Deductions in grade scores were based on blastomere asymmetry and increasing degrees of fragmentation.

All the oral progesterone was supplied by a single source. Each 200-mg oral capsule contained 200 mg of micronized progesterone United States Pharmacopeia and 140 mg of methocel E4M. After the two powders were titrated together using geometric dilution, they were placed in a size-zero gelatin capsule (Eli Lilly, Indianapolis, IN).

Progesterone assays were performed using the Immulyte system. The intra-assay and interassay coefficients of variation were 8.1%–13% and 6.9%–13%, respectively. The manufacturer of this progesterone assay system has not performed an evaluation of cross-reactivity with the most abundant metabolites of oral progesterone, 5α- and 5β-pregnanolone. We therefore tested for cross-reactivity by spiking 5-ng/mL progesterone standards with 30, 120, and 240 ng/mL of each of these two progestins.

Statistical comparisons were made using a standard software program. The Mann-Whitney U test was used for comparison of numbers and levels. Rates were compared using the χ^2 test, with the Yates' correction when necessary. For smaller groups, Fisher's exact test was used. A P value of $<.05$ was considered statistically significant.

RESULTS

Nineteen patients received IM progesterone and 24 patients received oral progesterone. There were no statistically significant differences between the two groups in mean age, response to stimulation, or retrieval outcome. Moreover, the number and quality of the embryos and the clinical pregnancy rate were not significantly different between the two groups (Tables 1 and 2). There were two miscarriages in the oral group, one of which was chromosomally abnormal, and no miscarriages in the IM group.

A statistically significant difference was observed in the implantation rate per embryo. The patients who received oral progesterone demonstrated a greater than twofold lower implantation rate per embryo compared with the patients who received IM progesterone (Table 2). This lower implantation rate was reflected in a lower multiple pregnancy rate in the oral group. We initially intended to enroll more patients; however, the study was terminated for ethical reasons because the differences in implantation rates were highly statistically significant.

TABLE 1

Intramuscular versus oral progesterone for IVF: comparison of stimulation data.

Variable	Route of administration	
	IM (n = 19)	PO (n = 24)
Age (y)	34.5 ± 0.57	34.9 ± .075
Day of hCG administration	11.9 ± .275	11.3 ± .502
E_2 level on day of hCG administration	1,964 ± 230.3	1,770 ± 173.2
No. of oocytes	15.8 ± 1.43	13.7 ± 1.10
No. of embryos	10.8 ± 1.20	10.1 ± .867
No. of embryos transferred	3.47 ± .193	3.46 ± .170
No. of highest-quality embryos	1.64 ± .095	1.45 ± .083
Mean embryo quality	1.86 ± .099	1.91 ± .072

Note: Values are means ± SEM. All differences were not statistically significant. PO = oral.

Overall, there was no difference in the circulating progesterone levels between the IM and oral groups. This remained true when we examined the subgroup of patients who were not pregnant and therefore did not have the enhanced progesterone level that accompanies a pregnancy. It was of interest, however, that on day 28, four patients in the oral group, none of whom were pregnant, had progesterone levels of <20 ng/mL (Table 3). Patients with E_2 levels of $>1{,}000$ pg/mL were given 5,000 U of hCG, and those with lower levels were given 10,000 U. There were no differences in progesterone levels, implantation rates, or pregnancy rates between the two dosage groups.

There was little cross-reactivity of 5α- and 5β-pregnanolone in our progesterone assay. The β compound was slightly more reactive than the α compound, but the maximum cross-reactivity was 1.3% (Table 4).

TABLE 2

Intramuscular versus oral progesterone for IVF: differences in pregnancy results.

Variable	Route of administration	
	IM (n = 19)	PO (n = 24)
No. of clinical pregnancies/no. of oocyte retrievals (%)	11/19 (57.9)	11/24 (45.8)
No. of patients with multiple implantation/total no. of pregnant patients (%)	9/11 (81.8)	4/11 (36.3)
No. of higher-order multiple implantations	4*	0
Implantation rate per embryo (%)	40.9	18.1†

Note: PO = oral.
* Three sets of quadruplets and one set of triplets.
† P = .004 (versus IM).

TABLE 3

Intramuscular versus oral progesterone for IVF: differences in circulating progesterone levels (mg/mL).

Variable	Route of administration	
	IM	PO
All patients		
No. of patients	19	24
Mean + SEM P4 level on day 21	77.6 ± 13.2	81.5 ± 16.2
Mean + SEM P4 level on day 28	76.3 ± 15.0	53.6 ± 10.1
Nonpregnant patients only		
No. of patients	8	13
Mean + SEM P4 level on day 21	54.3 ± 6.87	66.2 ± 19.63
Mean + SEM P4 level on day 28	28.5 ± 2.29	28.9 ± 5.06
No. of patients with P4 level of <20 ng/mL on day 21 (P4 levels of individual patients)	1 (19.0)	2 (10.8, 16.7)
No. of patients with P4 level of <20 ng/mL on day 28 (P4 levels of individual patients)	1 (18.0)	4 (5.2, 9.0, 9.1, 18.0)

Note: Differences between groups were not statistically significant.

DISCUSSION

This prospective randomized study demonstrated that oral progesterone is associated with a significantly lower implantation rate per embryo compared with IM progesterone when it is used for luteal support for IVF. This difference was observed without differences in circulating progesterone levels.

Over the past two decades, exogenous gonadotropins have been used to induce ovulation in infertile patients, and supplemental luteal progesterone has been used in an attempt to improve uterine receptivity in such cycles. The scientific foundation for progesterone use is that elevated luteal levels of estrogen decrease the incidence of embryo implantation, which is one proposed mechanism of postcoital hormonal contraception. Using the mouse model, it has been demonstrated clearly that increases in the progesterone-to-estrogen ratio negate the effects of estrogen alone on the endometrium and allow for implantation (1). Therefore, the high levels of E_2 that are produced during ovarian hyperstimulation may interfere with implantation unless supplemental progesterone is given.

Despite this theoretic benefit of luteal progesterone, and its wide use for such purposes, randomized studies have not shown that progesterone supplementation of gonadotropin-induced cycles (IVF or other) improves pregnancy rates in humans (2, 3). The situation is different, however, in gonadotropin treatment cycles that use GnRH pituitary down-regulation, because luteal phases become short (4) and IVF pregnancy rates are at least half as high as when progesterone supplements are not used (4, 5). Progesterone is necessary in this situation because GnRH agonists cause premature luteolysis (6, 7) by suppressing pituitary release of gonadotropins for up to 12 days after their discontinuation. In addition, GnRH agonists result in a decrease in the number of LH receptors found on granulosa cells, and they have the direct effect of suppressing granulosa cell E_2 and progesterone production (8).

The methods most often used to increase progesterone levels in IVF cycles include hCG administration and progesterone supplementation. Human chorionic gonadotropin increases both E_2 levels and progesterone levels, whereas progesterone has no effect on E_2 levels (9). However, both have been shown to be equally effective in supporting the luteal phase of patients undergoing IVF who are treated with a GnRH agonist, as measured by pregnancy rates (4).

More recently, oral micronized progesterone has been evaluated for use as luteal support (10). Preliminary information about luteal oral progesterone has been derived from studies that examined the use of oral progesterone in postmenopausal hormone replacement therapy (11). Substituting natural progesterone for the synthetic progestins commonly used in hormone therapy would have the advantage of avoiding the androgenic and psychotropic side effects that are associated with those medications. Oral progesterone initially was regarded as clinically ineffective because of poor intestinal absorption and rapid metabolism caused by the intestinal mucosa, intestinal flora, and a first-pass effect from the liver.

Micronizing progesterone (creating microspheres) involves processing the compound into a fine powder and suspending it in an oil carrier, increasing its bioavailability. Despite micronization, the intestinal absorption of oral progesterone is limited. Thus, the actions of the intestines and liver, coupled with limited absorption, result in a level of bioavailability that has been reported to be <10% (12). This is increased somewhat when the drug is taken with food (12).

Low circulating progesterone levels seem to be sufficient to counter the negative effects of estrogen replacement on the endometrium (13, 14). Histologic examination reveals

TABLE 4

Cross-reactivity of the Immulite progesterone assay with 5α- and 5β-pregnanolone.

Concentration added (ng/mL)	Progesterone assay cross-reactivity at 5 ng/mL	
	5α-pregnanolone (%)	5β-pregnanolone (%)
30	0.67	1.3
120	0.60	1.1
240	0.60	0.7

* A 0.67% rate of cross-reactivity means that 0.67% of 30 ng/mL of 5α-pregnanolone was measured as progesterone.

that the epithelium may not be converted to a secretory pattern; however, glandular cells do show mitotic arrest, and therefore hyperplasia is halted. Certainly, for infertility therapy, secretory conversion is desired. Therefore, increasing the dosage and decreasing the dosing interval should allow for adequate endometrial progesterone exposure.

When progesterone is given orally, progesterone levels have been reported to peak at approximately 2 hours, with a following half-life of approximately 2 hours (15). In addition, the absorption of oral progesterone shows considerable intersubject variability, so that some patients have high serum levels and others have low levels after taking the same dose (16). We attempted to overcome these potential problems by using a dosing interval of 8 hours, rather than the standard 12 hours, and by using a novel sustained-release methylcellulose vehicle. Methylcellulose forms a matrix around compounds that protects against stomach degradation, creating a sustained-release effect in the small intestine and aiding the absorption of the medication.

The use of oral progesterone for luteal support in patients undergoing IVF who are taking GnRH has been limited. A poor outcome was reported by Buvat et al. (17), who demonstrated that oral micronized progesterone in oil (100 mg at 8 A.M., 100 mg at noon, and 200 mg at 8 P.M.) produced a clinical pregnancy rate of 23% and an implantation rate per embryo of 7.5%, compared with rates of 45% and 19%, respectively, for IM progesterone (the difference was statistically significant). However, Pouly et al. (10) reported that oral progesterone (100 mg in the morning and 200 mg in the evening) resulted in a clinical pregnancy rate of 25% and an implantation rate of 29.9%, compared with rates of 28.8% and 35.3%, respectively, for vaginal progesterone gel (the difference was not statistically significant). The rates in our study are consistent with those of Pouly et al. (10).

Although there were overall differences in progesterone levels with the two routes of administration, and no differences in pregnancy rates, the number of patients in our study was too small to detect statistically significant differences in these areas. Overall, morning levels of progesterone were not different between the two groups, although the lowest levels were found in those patients who received the oral drug.

The rapid degradation of orally administered progesterone results in a high concentration of circulating metabolites, including deoxycorticosterone, estrone, and E_2. The most common metabolites, 5α and 5β reduced pregnanolones, circulate in concentrations that are higher than that of progesterone itself (15, 18). Earlier assays of progesterone have measured these two compounds, resulting in erroneous elevations of the perceived circulating progesterone levels. Our testing showed that these compounds were not detected by the Immulite assay system, so that we believe that we obtained an accurate impression of the levels of circulating progesterone.

We can only guess at the cause of oral progesterone's negative effects on embryo implantation. Overall, progesterone levels were not lower in the oral group; however, this group did contain the patients with the lowest progesterone levels. The progesterone metabolites, circulating at high levels, may bind to progesterone receptors and interfere with normal progesterone action by interfering with transcription cofactor or DNA binding. Alternatively, the 5α and 5β reduced pregnanolones are known to have high affinity for γ-aminobutyric acid receptors (19). Such receptors are present in the reproductive tract (20), and their activation may adversely affect pregnancy outcome.

Although we initially intended to enroll a larger number of patients in each group of the study, a disparity in the implantation rates led us to end the study prematurely for ethical reasons. The small number of patients lowers the power of this study and restricts our ability to comment on the usefulness of oral progesterone. Because there was a large difference between the two groups, however, we believe that we are justified in concluding that the difference in the implantation rate per embryo is significant when our protocols and method of administering oral micronized progesterone are used.

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