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Single and multidose pharmacokinetic study of a vaginal micronized progesterone insert (Endometrin) compared with vaginal gel in healthy reproductive-aged female subjects

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Objective: To determine pharmacokinetic profiles of two times a day and three times a day dosage regimens of Endometrin, a micronized progesterone vaginal insert for luteal support in assisted reproductive technology, compared with a gel.
Design: A single-center, randomized, open-label, single-day, and multiple-day (5 days) parallel design pharmacokinetic study.
Setting: University clinical research unit.
Patient(s): Three groups of six healthy subjects, ages 18 to 40 years.
Intervention(s): Endometrin vaginal inserts two times a day or three times a day, or gel daily.
Main Outcome Measure(s): Pharmacokinetic profiles.
Result(s): Progesterone serum concentrations increased rapidly following administration of Endometrin vaginal insert, producing higher peak concentrations (C_{max}) and clearing faster than gel. On the single day of dosing, mean C_{max} was 17.0 $\pm$ 2.7 ng/mL in the two times a day group, 19.8 $\pm$ 2.9 ng/mL in the three times a day group, and 6.82 $\pm$ 1.69 ng/mL in the gel group. Endometrin treatments reached steady state within the first 2 days (24–36 hours), much more rapidly than the gel, which had not reached steady state by 5 days. At 5 days, the Endometrin treatments produced sustained progesterone concentrations exceeding 10 mg/mL across 24 hours.
Conclusions: Endometrin vaginal inserts reached higher C_{max}, produced greater systemic exposure (area under the curve 0–24), achieved steady state more rapidly, and cleared more rapidly after termination of therapy than the comparator. (Fertil Steril® 2010;94:1296–301. ©2010 by American Society for Reproductive Medicine.)

Key Words: Endometrin, progesterone, progesterone supplementation, luteal support, assisted reproductive technology, vaginal, pharmacokinetics, PK, in vitro fertilization, IVF

In vitro fertilization and other assisted reproductive technology (ART) treatments often require exogenous luteal phase support (1–3). Normal, adequate endometrial preparation includes endometrial proliferation by estrogen, and secretory transformation by progesterone, whether provided endogenously or exogenously. Available exogenous progesterone options for endometrial support in ART cycles currently include oral, intramuscular (IM), and, more recently, vaginal formulations.

Although the convenience of oral administration is attractive, progesterone is rapidly cleared by first-pass hepatic metabolism after oral administration (4–6). Consequently, systemic bioavailability of progesterone is signficantly lower after oral than after vaginal administration (7). To compensate for this low bioavailability, oral administration requires high doses that are associated with systemic adverse effects, including drowsiness, flushing, and nausea (4).

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In addition, the pharmacokinetic properties of orally administered progesterone are influenced by food intake (8). Furthermore, orally administered progesterone has been shown to have limited efficacy in inducing an in-phase secretory endometrium, and evidence indicates a less favorable ART outcome using oral application compared with either the IM or vaginal route (1, 9, 10).

Progesterone is rapidly absorbed after IM administration; high plasma concentrations are generally achieved within 2 hours, and peak concentrations within 8 hours (4, 8). Nevertheless, the IM administration of the oil-based product can be extremely uncomfortable. Daily injections are required to maintain adequate serum progesterone concentrations, especially in patients with ovarian failure who lack any endogenous contribution. Reaching placental autonomy may take up to 10 to 12 weeks, thus necessitating a protracted use of daily IM injections. Injections can lead to patient discomfort, inflammatory reactions at the injection site, sterile abscesses, and possible infections (4). Rare severe complications may include hematomas and secondary nerve compression (11).

The vaginal route has recently gained attention because it avoids the variable absorption and high first-pass hepatic metabolism after oral ingestion, while also preventing the uncomfortable, often painful, IM injection (4). Vaginal application can result in sustained plasma concentrations (12–14). Experimental and clinical data also suggest that vaginal application provides significant levels of

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progesterone to the endometrial tissue, inducing secretory transformation (4, 6, 15, 16).

Progesterone is available for vaginal administration (17–24). Its pharmacokinetics compare favorably with oral administration. Progesterone given vaginally results in greater bioavailability and less variability in serum concentrations than does oral progesterone. Thus, vaginal administration of progesterone provides a more reliable delivery of progesterone than oral administration. Furthermore, a cohort observational study comparing vaginal administration with IM injection found equivalent endometrial development and pregnancy rates (18).

A novel vaginal micronized progesterone insert (Endometrin) has recently been approved for luteal support in the treatment of infertile women undergoing ART (25). The comparative pharmacokinetic parameters of this new vaginal insert and a previously marketed vaginal gel progesterone formulation had not been determined. The purpose of this study was to determine the pharmacokinetic and safety profiles of two dosage regimens of the micronized progesterone 100 mg vaginal insert (twice a day and three times a day) compared with the pharmacokinetic and safety profile of the 8% vaginal gel (90 mg every day) in normal, reproductive-aged females with an intact uterus. The principal pharmacokinetic objectives of this study were to: [1] obtain single-day and steady-state progesterone pharmacokinetics for the three treatment groups, [2] describe progesterone steady-state pharmacokinetics, and [3] compare progesterone pharmacokinetics among the three treatment groups.

MATERIALS AND METHODS

The study protocol was approved by and conducted in accordance with the guidelines of the University of Miami Human Subjects Research Committee (institutional review board); it was also conducted in accordance with Good Clinical Practices and International Council on Harmonization guidelines. Study personnel obtained written informed consent directly from all subjects before their entry into the study. All clinical study procedures were conducted in the Phase I Clinical Research Unit of the Division of Clinical Pharmacology of the University of Miami.

Study Design

This was a single-center, randomized, open-label, parallel design pharmacokinetic study in normally cycling female subjects between 18 and 40 years of age with an intact uterus. Eighteen eligible subjects (six per treatment group) were randomly assigned to receive one of two different dosing regimens of a 100 mg vaginal insert (two or three times a day) (Endometrin, Ferring Pharmaceuticals, Inc., Parsippany, NJ) or 8% gel (90 mg every day) (Crinone, Serono, Inc., Rockland, MA). The study was divided into four phases: screening, single-day (single day of dosing), washout, and multiple day (5 days of dosing). Blood samples for pharmacokinetic analyses were collected over a 48-hour period during the single-day phase and starting on day 5 of the multiple-day phase. A 7-day washout phase, immediately following the 24 hours of the single-day administration, separated the single-day and multiple-day phases of the study.

Eligible subjects were in general good health as determined by medical history and physical examination. Subjects were required to have regular menstrual cycles (24 to 35 days), a body mass index of 18 to 28 kg/m^2, a documented negative Pap smear, and a negative urine pregnancy test. After successful screening, the subject was randomly assigned to one of three treatment arms of study medication, initiated after menstrual flow ceased, between cycle days 5 and 8 of the subject's menstrual cycle. During the single-day phase, study drug was administered for 1 day (a single dose for the every day treatment, two doses 12 hours apart for the two times a day treatment, and three doses 8 hours apart for the three times a day treatment). Each dose of study medication was administered under the direct observation of a member of the study team. Blood samples for pharmacokinetic analysis were obtained pre-dose (0 hour), and at 2, 4, 8, 12, 16, 24, 36, and 48 hours following the first dose of study medication.

After the 7-day washout phase, the subject returned to the phase I inpatient unit for a stay of approximately 6 overnights and received 5 days of treatment during the multiple-day phase. On day 5 of medication, blood samples for pharmacokinetic analysis were obtained from all subjects pre-dose (0 hour) and at 2, 4, 8, 12, 16, 24, 36, and 48 hours after first dosing. For the vaginal insert two times a day or three times a day regimens, medication was administered immediately after the 12-hour or immediately after the 8- and 16-hour blood samples, respectively. All subjects were assessed for the following safety parameters during participation in the study: adverse events (AEs), serious AEs (SAEs), clinical laboratory evaluations (hematology and serum chemistry), vital signs (blood pressure, heart rate, and body temperature), and electrocardiogram.

Analytical Methods

A validated, sensitive, and specific radioimmunoassay using an ^{125}I labeled progesterone derivative was used to quantify progesterone concentrations in human serum. Intraassay coefficients of variation for quality control samples ranged from 2% to 8%, and interassay coefficients of variation for quality control samples ranged from 5% to 10%.

Statistical Methods and Pharmacokinetic Analysis

Treatment group comparisons for demographic and baseline characteristics were based on Fisher's exact test for qualitative variables and analysis of variance or the Kruskal-Wallis test, as appropriate, for quantitative variables. Summary statistics were tabulated by treatment group.

Pharmacokinetic parameters determined for single-day and for multiple-dose, multiple-day, treatment day 5 included maximum observed serum concentration (C_{max}), time to maximum observed serum concentration (T_{max}), area under the serum concentration–time curve over the dosing interval (AUC_{0-T}) and for the 24-hour period (AUC_{0-24}). Trough (predose) concentrations were used to assess the onset of steady state. The Fluctuation Index over a 24-hour period was calculated as the $(C_{max} - C_{min})/(AUC_{(0-24)}/24)$, where C_{min} was determined by inspection. Pharmacokinetic calculations were done using built-in functions in the Excel 2003 component of Microsoft Office 2003 (Microsoft, Redmond, WA) and confirmed by comparison to output from WinNonlin 4.0.1 (Pharsight Corporation, Sunnyvale, CA).

RESULTS

A total of 18 subjects were screened and randomized to treatment. Six subjects were assigned to each treatment group (Table 1). All subjects were female, and ranged in age from 18 to 40 years. Most of the subjects were Hispanic (13/18, 72%). Body mass index ranged from 22 to 28 kg/m^2. No statistically significant differences were observed in demographic characteristics across treatment groups or between the groups. Gynecologic history was generally similar across treatment groups. A statistically significant difference was observed across groups in average cycle length (P=.008), but all cycle lengths were normal and the difference was not considered clinically meaningful. All 18 subjects completed both the single day of dosing and multiple days of dosing. Compliance was 100% in all groups.

Pharmacokinetic Results

Single-day Treatment

The mean concentration–time profiles for the three treatment groups during the single-day phase are presented in Figure 1. Figure 1 demonstrates consistently higher progesterone concentrations for both the two and three times a day Endometrin insert groups than for the gel group. The mean progesterone concentrations observed during the first 8 hours after administration of the first dose (hours 0–8 in the single-day profile) were nearly identical between the two and the three times a day vaginal insert groups. The

TABLE 1

Subject demographic characteristics and gynecological history.

Characteristic	Insert 100 mg bid (n = 6)	Insert 100 mg tid (n = 6)	Gel 90 mg qd (n = 6)	Insert Combined (n = 12)	*P* value
Race					
Hispanic	5 (83%)	3 (50%)	5 (83%)	8 (67%)	Across: .527[a] Insert combined versus Gel: .615[a]
Caucasian	1 (17%)	3 (50%)	1 (17%)	4 (33%)	
Age (y)					
Mean (SD)	35.8 (4.96)	32.7 (9.63)	35.5 (2.59)	34.3 (7.48)	Across: .652[b] Insert combined versus Gel: .700[c]
Minimum, maximum	29, 40	18, 40	33, 39	18, 40	
BMI (kg/m^2)					
Mean (SD)	24.7 (2.73)	26.0 (1.97)	26.8 (1.17)	25.3 (2.38)	Across: .218[b] Insert combined versus Gel: .168[c]
Minimum, maximum	22, 28	23, 28	25, 28	22, 28	
No. of pregnancies					
Mean (SD)	2.2 (1.47)	1.3 (1.51)	1.2 (0.75)	1.8 (1.48)	Across: .380[b] Insert combined versus Gel: .383[c]
Minimum, maximum	1, 5	0, 3	0, 2	0, 5	
No. of births					
Mean (SD)	2.2 (1.47)	1.0 (1.26)	0.8 (0.75)	1.6 (1.44)	Across: 0.146[b] Insert combined versus Gel: .254[c]
Minimum, maximum	1, 5	0, 3	0, 2	0, 5	
No. of abortions					
Mean (SD)	0.0 (0.00)	0.3 (0.52)	0.3 (0.82)	0.2 (0.39)	Across: .505[b] Insert combined versus Gel: .559[c]
Minimum, maximum	0, 0	0, 1	0, 2	0, 1	
Average cycle length					
Mean (SD)	27.0 (1.55)	29.3 (1.03)	26.5 (1.64)	28.2 (1.75)	Across: .008[b,*] Insert combined versus Gel: .070[c]
Minimum, maximum	25, 28	28, 30	25, 28	25, 30	

Note: SD = standard deviation; BMI = body mass index; Insert = Endometrin; Gel = 8% Crinone; bid = two times a day; tid = three times a day; qd = every day.

* Statistically significant difference ($P \leq .05$).

[a] *P* value from two-tailed Fisher's exact test.

[b] *P* value from one-way analysis of variance or Kruskal-Wallis test, as appropriate.

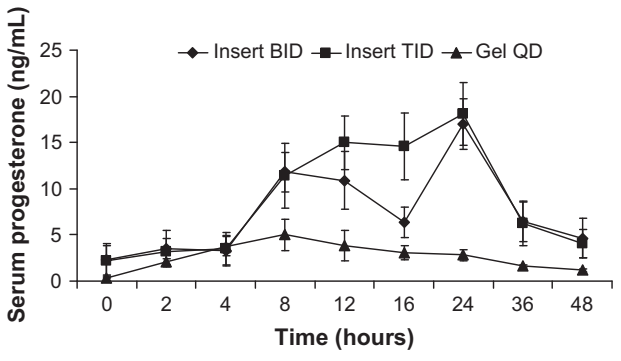
[c] *P* value from *t* test or Wilcoxon Rank Sum test, as appropriate.

Blake. Endometrin vaginal progesterone PK study. Fertil Steril 2010.

concentrations in the three times a day group exceeded those of the two times a day group beyond 8 hours, the time that a second dose was administered in the three time a day group. Derived pharmacokinetic parameters for serum progesterone are presented in Table 2. On the single day of dosing, mean C_{max} was 17 ng/mL in the two times a day insert group, 19.8 ng/mL in the three times a day insert group, and 6.8 ng/mL in the vaginal gel group. The 24-hour systemic exposure AUC_{0-24} was 217 ng•h/mL in the two times a day insert group, 284 ng•h/mL in the three times a day insert group, and 81 ng•h/mL in the gel group.

FIGURE 1

Single-day treatment phase: mean (±SEM) serum progesterone concentrations (ng/mL).



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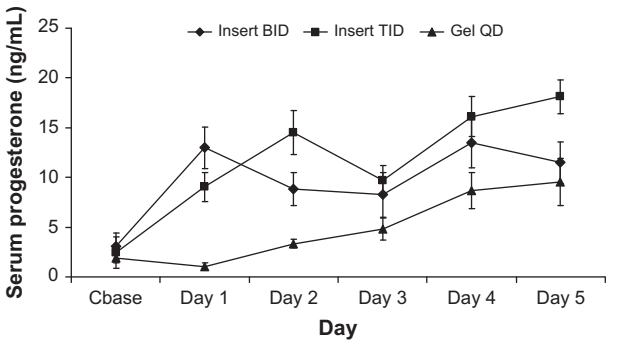
Approach to Steady State

The kinetics of the approach to steady state were monitored by sampling trough (predose) concentrations once per day during the multiple days of dosing. The mean progesterone trough concentrations each day during the multiple-day phase are presented graphically in Figure 2. Figure 2 demonstrates that the progesterone concentrations produced with the vaginal insert two times a day and the vaginal insert three times a day dosing approximated steady state progesterone concentrations within 24 to 32 hours after the first dose. The approach to steady state with the vaginal gel was considerably slower. The trough concentrations for the gel every day group did not fully reach steady state by the end of the fifth day of dosing.

Trough levels first exceeded 10 ng/mL on day 1 in the two times a day insert group and on day 2 in the three times a day insert group, but had not reached 10 ng/mL even by the end of day 5 in the gel group (Fig. 2). These results suggest that the vaginal insert two and three times a day can reach a targeted concentration range in a substantially shorter time than the vaginal gel therefore earlier in the course of a treatment program.

FIGURE 2

Days 1–5 of the multiple-day treatment phase: mean (±SEM) serum progesterone trough concentrations (ng/mL).



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Multiple-day Treatment: day 5

Figure 3 provides a display of the day 5 mean serum progesterone concentrations for the three treatment groups. Both insert treatments were associated with sustained concentrations above 10 ng/mL throughout the 24-hour period of day 5, a progesterone threshold associated with the midluteal phase, whereas the gel treatment did not continuously maintain this level. The derived pharmacokinetic parameters for the three treatment regimens are displayed in Table 2. The vaginal insert two and three times a day groups demonstrated a higher C_{max} than the vaginal gel group. The insert regimens provided greater systemic exposures than did the gel as measured by the $AUC_{0–24}$, with values on day 5 ranging from 264 ng•h/mL (gel) to 436 ng•h/mL (insert three times a day).

Comparing the two and three times a day Endometrin regimens revealed that the three times a day regimen, with its 50% higher daily dose, produced higher systemic concentrations, although only about 30% higher ($AUC_{0–24}$: 327 ng•h/mL vs. 436 ng•h/mL). The less than dose–proportional response may have been a result of random variability, but it is also consistent with a less than dose–proportional response seen across most treatments in a previous study with Endometrin (Ferring data on file). Comparison of Figures 1 and 3 demonstrates that the 8% vaginal gel administered every day showed substantial accumulation between day 1 and day 5 of treatment. C_{max} on day 5 (14.3 ng/mL) was more than twice the C_{max} observed after a single application of vaginal gel (6.8 ng/mL), evidence of the slow approach to steady state.

The three treatment regimens differed with respect to their variability in the pharmacokinetic parameters. The vaginal gel had the greatest between-subject variability in its day 5 pharmacokinetic parameters, whereas the vaginal insert three times a day had the least variability. The coefficient of variation for C_{max} on day 5 in the gel group was 39.7%, but only 23.2% in the Endometrin three times a day group. It was intermediate, 29.9%, in the Endometrin two times a day group.

The decay phases for both Endometrin two and three times a day were almost identical at 24 and 48 hours postdose. The gel had a prolonged elimination phase, in which concentrations decreased by only about a factor of four during the 48-hour observation period. The elimination phase slowed even further with repeated dosing, such that, by the end of the 5-day period, there was essentially no decay in serum progesterone concentration over the 36-hour window from 12 hours to 48 hours postdose.

Safety Results

An AE is defined as any untoward medical occurrence in a trial subject who has received any study medication; the event does not necessarily have a causal relationship with the study medication. Serious adverse events encompass the following events: death, life-threatening (i.e., at immediate risk of death), in-patient hospitalization or prolongation of existing hospitalization, persistent or significant disability/incapacity, congenital anomaly/birth defect.

A total of seven AEs were reported during the study, and no SAEs. One subject in the vaginal gel every day group reported headache. All other AEs occurred in the three times a day group. Three subjects experienced mild vaginal bleeding, which was judged as probably related to study drug. For all three subjects, the mild bleeding occurred during the washout phase following the single day of

TABLE 2

Mean (±SEM) serum progesterone pharmacokinetic parameters.

		Endometrin 100 mg bid (n=6) Mean ± SEM	Endometrin 100 mg tid (n=6) Mean ± SEM	Gel 90 mg qd (n=6) Mean ± SEM
			Single-day treatment	
C_{max}	ng/mL	17.0 ± 2.7	19.8 ± 2.9	6.8 ± 1.69
T_{max}	h	24.0 ± 0.0	17.3 ± 3.0	13.3 ± 2.5
AUC (0–T)	ng•h/mL	88.4 ± 21.1	41.7 ± 15.5	80.9 ± 17.0
AUC (0–24)	ng•h/mL	217 ± 46	284 ± 58	81 ± 17.0
			Multiple-dose, multiple-day treatment, day 5	
C_{max}	ng/mL	18.5 ± 2.3	24.1 ± 2.3	14.3 ± 2.3
T_{max}	h	18.0 ± 3.8	18.0 ± 3.8	12.3 ± 5.2
C_{min}	ng/mL	8.9 ± 1.85	10.9 ± 2.7	7.4 ± 1.43
AUC (0–T)	ng•h/mL	167 ± 24	127 ± 14	264 ± 46
AUC (0–24)	ng•h/mL	327 ± 52	436 ± 43	264 ± 46
Cl/F	L/h	657 ± 87	846 ± 112	417 ± 95

Note: bid = two times a day; tid = three times a day; qd = every day; AUC = area under the curve.

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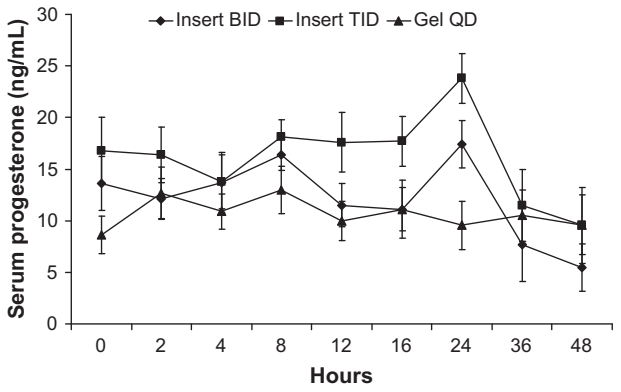
FIGURE 3

Day 5 of the multiple-day treatment phase: mean (±SEM) serum progesterone concentrations (ng/mL).



Blake. Endometrin vaginal progesterone PK study. Fertil Steril 2010.

three times a day dosing, and none had episodes of bleeding while still on the progesterone treatment. This is consistent with normal withdrawal bleeding following progesterone exposure, rather than breakthrough bleeding. One subject each experienced abdominal pain, back pain, and rash. No subjects in the two times a day or gel groups reported vaginal bleeding during the study. All AEs were mild in intensity. All events resolved without treatment within 3 to 4 days. No deaths, SAEs, or AEs that led to withdrawal of study drug were reported during the study.

DISCUSSION

This article describes a single-center, randomized, open-label, pharmacokinetic study of Endometrin vaginal inserts in 18 reproductive-aged cycling female subjects between 18 and 40 years of age with an intact uterus. The primary objective of this randomized clinical trial was to determine the pharmacokinetic profiles of two dosage regimens of the vaginal micronized progesterone insert, Endometrin (100 mg two times a day and 100 mg three times a day) compared with the pharmacokinetic profile of the available 8% vaginal gel (90 mg every day) in healthy, reproductive-aged females. The study was designed to compare single-day and steady-state progesterone pharmacokinetics for the three treatment groups, describe progesterone steady-state pharmacokinetics, and compare progesterone pharmacokinetics among the three treatment groups.

This study suggests that the pharmacokinetic profile of both dosage regimens of the vaginal insert formulation of progesterone compares favorably with the vaginal gel formulation. In this clinical trial, vaginal insert administration of progesterone was associated with a rapid rise in progesterone concentrations, significant plasma concentrations and bioavailability, a relatively rapid achievement of steady-state concentration, and less variability of plasma progesterone concentration than the gel formulation. Progesterone serum concentrations increased rapidly following the administration of the vaginal insert and produced higher peak concentrations than did the vaginal gel. On the single day of dosing, mean C_{max} was 17 ng/mL in the two times a day insert group and 19.8 ng/mL in the three times a day insert group, but only 6.8 ng/mL in the vaginal gel group. The vaginal insert treatments also appeared to reach steady-state concentrations more rapidly than did the vaginal gel, based upon trough concentrations. The progesterone concentrations produced by the insert approximated the steady state progesterone concentrations by 24 to 32 hours after the start of dosing. The vaginal gel was estimated to require 6 or more days to reach the steady state serum concentrations.

Because a midluteal phase serum progesterone level of 10 ng/mL is associated with adequate corpus luteum function and results in endometrium that is capable of maintaining pregnancy (26–29), it is important to note that both the two and the three times a day vaginal insert regimens provided adequate progesterone concentrations, with serum levels exceeding 10 ng/mL early in the multiple-day treatment phase, whereas the vaginal gel required nearly 1 week to reach the targeted physiologic level. In addition, progesterone concentrations were sustained in excess of 10 ng/mL throughout the 24-hour interval of day 5 of the multiple-day dosing phase with both insert regimens. The gel had the greatest variability in pharmacokinetic parameters, whereas the insert three times a day had the least variability.

Progesterone levels fell much more precipitously after discontinuation of either insert dosage (two or three times a day) than following withdrawal of treatment by gel.

Both the vaginal inserts and the vaginal gel were generally safe and well tolerated. All AEs were mild in intensity and resolved without treatment within 3 to 4 days. No deaths, SAEs, or AEs that led to withdrawal of study drug were reported during the study.

Limitations of our study include the small sample size and considerable variability of plasma concentrations among the study subjects. We compared two and three times a day regimens to the every day regimen of the vaginal gel. Thus, this is not a direct comparison of single-dose pharmacokinetics between the two formulations. Nevertheless, the dosages and frequencies of administration correspond to those used in clinical practice. Endometrin provides circulating progesterone levels that are easily measurable, thus allowing clinical assessment of the adequacy of exposure for individual patients in a clinical setting. Although individual variation in progesterone levels is great, and the predictive value of progesterone measurements is limited, those clinicians who want to measure progesterone levels for their treatment paradigm can easily do so.

Patients were randomized, and medication was started as soon as possible in the follicular phase (waiting until menstrual flow would not interfere with drug absorption), but because menstrual cycles were not controlled, the need for an adequate washout period between the single- and multiple-day exposure may have pushed some subjects into the periovulatory period during the multiple-day phase. The intention in starting all subjects in the early follicular phase was to allow a 7-day washout period without having the subjects ovulate or enter the luteal phase (expected days 7–11) before the second sampling. However, we also recognized that, with human variability, some women might indeed have ovulated before or during the multiple-day portion of the study. Estradiol and progesterone levels were obtained before initiation of treatment in both the single-day and multiple-day segments of this trial. We sampled blood to identify those women likely to be in the periovulatory/luteal phase of their cycle.

Ovulation would be expected to increase measured serum progesterone levels by adding endogenous to exogenous progesterone. Some women in each treatment group probably ovulated, given their initial progesterone levels of about 2 ng/mL or greater at the start of the multiple-day segment. Although initial progesterone levels were higher at baseline (time = 0) for these possibly ovulatory women, at almost all further time points across the 5 days of sampling, the

mean progesterone levels were actually higher when these subjects were excluded from analyses, and lower when they were included. Thus, even though we might expect endogenous progesterone to mask the levels achieved by exogenous administration, this was not the result observed. Therefore, we feel confident that we are not crediting the exogenous treatment with progesterone levels that may have been elevated by an endogenous contribution. So, because we had no reason to exclude these possibly ovulatory subjects from the overall evaluation, we have included them in the analysis.

Because of the unfavorable pharmacokinetics and the limited ability to achieve sustained serum progesterone concentrations with the oral administration of progesterone (1) and the discomfort and potential complications of repeated IM injections (4, 11), recent investigative and developmental efforts have been aimed at achieving successful alternative means of delivery of progesterone. The vaginal route avoids the variable absorption and high first-pass hepatic metabolism after oral ingestion and also prevents the uncomfortable and sometimes painful IM injection (6). Vaginal application can result in sustained plasma concentrations; experimental and clinical data suggest that the vaginal route is associated with significant uterine levels of progesterone and rates of secretory transformation (4, 6, 15, 16). Furthermore, several studies have demonstrated that vaginal application can result in sustained plasma concentrations suitable for IVF protocols (1).

This study demonstrates that two dosage regimens of a novel vaginal insert formulation of progesterone can achieve relatively high serum progesterone concentrations, reach steady state within 24 to 32 hours, and maintain mean concentrations above 10 ng/mL. Endometrin vaginal insert formulations reached higher C_{max}, produced greater systemic exposure (AUC_{0-24}), achieved steady state more rapidly, and cleared more rapidly after termination of therapy than the comparator. Although this study focuses on the pharmacokinetic characteristics of Endometrin, a large clinical trial demonstrated its efficacy (25).

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REFERENCES

1. Toner JP. The luteal phase: luteal support protocols. In: Gardner DK, Weissmann A, Howles CM, Shohan Z, eds. Textbook of assisted reproductive techniques. London: Taylor & Francis, 2004:639–50.
2. Pabuccu R, Akar ME. Luteal phase support in assisted reproductive technology. Curr Opin Obstet Gynecol 2005;17:277–81.
3. Penzias AS. Luteal phase support. Fertil Steril 2002;77:318–23.
4. Tavaniotou A, Smitz J, Bourgain C, Devroey P. Comparison between different routes of progesterone administration as luteal phase support in infertility treatments. Hum Reprod Update 2000;6:139–48.
5. Whitehead MI, Townsend PT, Gill DK, Collins WP, Campbell S. Absorption and metabolism of oral progesterone. Br Med J 1980;280:825–7.
6. Bulletti C, de Ziegler D, Flamigni C, Giacomucci E, Polli V, Bolelli G, et al. Targeted drug delivery in gynaecology: the first uterine pass effect. Hum Reprod 1997;12:1073–9.
7. Levine H, Watson N. Comparison of the pharmacokinetics of Crinone™ 8% administered vaginally versus Prometrium administered orally in postmenopausal women. Fertil Steril 2000;73:516–21.
8. Simon JA, Robinson DE, Andrews MC, Hildebrand JR 3rd, Rocci ML Jr, Blake RE, et al. The absorption of oral micronized progesterone: the effect of food, dose proportionality, and comparison with intramuscular progesterone. Fertil Steril 1993;60:26–33.
9. Lane G, Siddle NC, Ryder TA, Pryse-Davies J, King RJ, Whitehead MI. Dose dependant effects of oral progesterone on the oestrogenised postmenopausal endometrium. Br Med J (Clin Res Ed) 1983;287:1241–5.
10. Devroey P, Braekmans P, Camus M, Khan I, Smitz J, Staessen C, et al. Embryo donation in patients with primary ovarian failure. Hum Reprod 1988;3(Suppl 2):85–7.
11. Kuhle JW, Swoboda B. Gluteal compartment syndrome after intramuscular gluteal injection. Z Orthop Ihre Grenzgeb 1999;137:366–7.
12. Kimzey LM, Gumowski J, Merriam GR, Grimes GJ Jr, Nelson LM. Absorption of micronized progesterone from a nonliquefying vaginal cream. Fertil Steril 1991;56:995–6.
13. Archer DF, Fahy GE, Viniegra-Sibal A, Anderson FD, Snipes W, Foldesy RG. Initial and steady-state pharmacokinetics of a vaginally administered formulation of progesterone. Am J Obstet Gynecol 1995;173:471–8.
14. Norman TR, Morse CA, Dennerstein L. Comparative bioavailability of oral and vaginally administered progesterone. Fertil Steril 1991;56:1034–9.
15. Cicinelli E, de Ziegler D, Buletti C, Matteo MG, Schonauer LM, Galantino P. Direct transport of progesterone from vagina to uterus. Obstet Gynecol 2000;95:403–6.
16. Miles RA, Paulson RJ, Lobo RA, Press MF, Dahmoush L, Sauer MV. Pharmacokinetics and endometrial tissue levels of progesterone after administration by intramuscular and vaginal routes: a comparative study. Fertil Steril 1994;62:485–90.
17. Jobanputra K, Toner JP, Denoncourt R, Gibbons WE. Crinone 8% (90 mg) given once daily for progesterone replacement therapy in donor egg cycles. Fertil Steril 1999;72:980–4.
18. Gibbons WE, Toner JP, Hamacher P, Kolm P. Experience with a novel vaginal progesterone preparation in a donor oocyte program. Fertil Steril 1998;69:96–101.
19. Chantilis SJ, Zeitoun KM, Patel SI, John DA, Madziar VA, McIntire DD. Use of Crinone vaginal progesterone gel for luteal support in in vitro fertilization cycles. Fertil Steril 1999;72:823–9.
20. Schoolcraft WB, Hesla JS, Gee MJ. Experience with progesterone gel for luteal support in a highly successful IVF programme. Hum Reprod 2000;15:1284–8.
21. Propst AM, Hill JA, Ginsburg ES, Hurwitz S, Politch J, Yanushpolsky EH. A randomized study comparing Crinone 8% and intramuscular progesterone supplementation in in vitro fertilization–embryo transfer cycles. Fertil Steril 2001;76:1144–9.
22. Kleinstein J. Efficacy and tolerability of vaginal progesterone capsules (Utrogest ™ 200) compared with progesterone gel (Crinone ™ 8%) for luteal phase support during assisted reproduction. Fertil Steril 2005;83:1641–9.
23. Levine H. Luteal support in IVF using the novel vaginal progesterone gel Crinone 8%: results of an open-label trial in 1184 women from 16 U.S. centers. Fertil Steril 2000;74:836–7.
24. Fanchin R, de Ziegler D, Bergeron C, Righini C, Torrisi C, Frydman R. Transvaginal administration of progesterone. Obstet Gynecol 1997;90:396–401.
25. Doody KJ, Schnell VL, Foulk RA, Miller CE, Kolb BA, Blake EJ, Yankov VI. Endometrin for luteal phase support in a randomized, controlled, open-label, prospective in-vitro fertilization trial using a combination of Menopur and Bravelle for controlled ovarian hyperstimulation. Fertil Steril 2009;91:1012–7.
26. Speroff L, Fritz MA. Clinical gynecologic endocrinology and infertility. 7th ed. Philadelphia, PA: Lippincott, Williams & Wilkins, 2005:260.
27. Csapo AL, Pulkkinen MO, Wiest WG. Effects of lutectomy and progesterone replacement in early pregnant patients. Am J Obstet Gynecol 1973;115:759–65.
28. Schneider MA, Davies MC, Honour JW. The timing of placental competence in pregnancy after oocyte donation. Fertil Steril 1993;59:1059–64.
29. Azuma K, Calderon I, Besanko M, Maclachlan V, Healy D. Is the luteo-placental shift a myth? Analysis of low progesterone levels in successful ART pregnancies. J Clin Endocrinol Metab 1993;77:195–8.