Symposium: Embryo implantation failure and recurrent miscarriage

Progesterone supplementation to prevent recurrent miscarriage and to reduce implantation failure in assisted reproduction cycles

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Abstract

Implantation failure has been questioned for many cases of recurrent miscarriage and unsuccessful assisted reproduction. The exact cause of implantation failure is not known, but luteal phase defect is encountered in many of these cases. Consequently, women with recurrent miscarriages have been treated with progesterone supplementation with various degrees of success, and a recent meta-analysis has shown trends for improved live birth rates in those women. Progesterone probably acts as an immunological suppressant blocking T-helper (Th1) activity and inducing release of Th2 cytokines. Numerous studies have confirmed that ovarian stimulation used in assisted reproduction is associated with luteal phase insufficiency, even when gonadotrophin-releasing hormone antagonists are used. In those patients, advanced endometrial histological maturity and a decrease in the concentration of cytoplasmic progesterone receptors are observed. Progesterone supplementation results in a trend towards improved ongoing and clinical pregnancy rates, except in patients treated with human menopausal gonadotrophin-only regimens, in whom ongoing pregnancy rates increase significantly. More randomized controlled trials are needed to increase the power of the currently available meta-analyses to further evaluate progesterone supplementation in both conditions.

Keywords: assisted reproduction technology, implantation failure, maternal immune response, ovarian stimulation, progesterone, recurrent miscarriage
**Introduction**

Sir Peter Medawar, the Nobel prize laureate of 1960, described implantation as an immunological paradox whereby the semiallograft human conceptus, immunologically foreign to the mother, evades immune rejection (Medawar, 1961). Since then, numerous detailed studies have shown that implantation is a two-phase process. The first phase involves the apposition and adhesion of the embryo to the endometrial epithelium, a process largely mediated by integrins, mucins, trophinins and tastins. The second, or invasive, phase consists of two steps: early and deep invasion. Early invasion is an interplay between matrix metalloproteases (MMP) secreted by the embryo and tissue inhibitors of these proteases (TIMMP) secreted by the endometrium. Deep invasion is an interplay between T-helper (Th1) cytokines preventing implantation and Th2 cytokines enhancing implantation.

Excessive implantation is thought to result in pathological adhesion of the placenta (i.e. placenta accreta), while implantation failure has been blamed for many cases of recurrent miscarriage, recurrent failure of assisted reproduction and even pre-eclamptic toxaemia (Edwards et al., 1995).

For many years, progesterone therapy has been used to reduce implantation failure. The aim of this manuscript was to appraise the available literature and review the rationale for the use of progestational agents in these cases.

**Role of progesterone in implantation**

Morphological and physiological changes of the endometrium are triggered by oestrogens and progesterone secreted cyclically by the ovary. It is established that these cyclical modifications allow the creation of a suitable endometrial environment for embryo implantation and maintenance of early pregnancy. Progesterone-receptor antagonists induce abortion if given within the first 7 weeks of pregnancy (Peyron et al., 1993). Similarly, surgical removal of the ovary with the corpus luteum of pregnancy before 8 weeks of gestation leads to spontaneous miscarriage (Csapo and Pulkkinen, 1978).

A decrease in the amount or the duration of progesterone secretion by the corpus luteum, or the lack of an adequate response by the endometrium, results in luteal phase deficiency (Jones, 1991; Ginsburg, 1992) and subsequent pregnancy failure (Porter and Scott, 2005). Luteal phase defect (LPD) has been considered a cause of poor reproductive performance for over 50 years (Jones, 1949). Since then, the preferred method to diagnose LPD has been the morphological examination of a precisely timed luteal phase endometrial biopsy according to Noyes’ criteria (Noyes et al., 1950). The endometrium is considered ‘out of phase’ when, based on morphological criteria, it lags 2 days behind the actual ovulation date estimated by counting backward from the date of onset of the next menstrual period. Traditionally, the diagnosis of LPD is made if two consecutive biopsies are found to be out of phase (Wentz, 1980). There have been multiple reports of significant inter- and intraobserver variability in the histological evaluation for dating of the human endometrium (Li et al., 1989; Myers et al., 2004). Recently, the need and usefulness of histological dating of the endometrium, but not the existence of the diagnosis of LPD, has been questioned (Coutifaris et al., 2004). These observations indicate that the timed endometrial biopsy and histological dating of the endometrium provide no additional clinical benefit in the routine evaluation of infertility.

As recently reviewed by Check (2002), there may well be two types of LPD: one related to the presence of immature follicles, and one where the follicles are mature. In both types, supplemental therapy with progesterone is effective in creating a healthy uterine environment.

Despite the absence of robust evidence to support LPD as a cause of recurrent pregnancy loss, progestational agents have been commonly prescribed to prevent miscarriage in the first trimester. Early studies (Tho et al., 1983; Daya et al., 1988) showed that women who suffer miscarriages benefit from progesterone supplementation, and a recent meta-analysis found a trend for improved live birth rates in these women (Oates-Whitehead et al., 2003).

The practice of prescribing luteal phase support in long protocol IVF cycles is widespread among fertility physicians (Ludwig and Diedrich, 2001; Toner, 2001). An earlier meta-analysis (Soliman et al., 1994) showed that progesterone supplementation is of clinical value in the maintenance of early pregnancies achieved after IVF. The rationale is that pituitary desensitization with gonadotrophin-releasing hormone (GnRH) agonist causes an inadequate function and lifespan of the corpus luteum (Smitz et al., 1988).

Studies have shown differences in expression of endometrial molecules and leukocytes in the peri-implantation period in women with implantation failure and recurrent miscarriage. The role of progesterone in improving reproductive performance is probably through immune modulation (Choi et al., 2000).

**Progesterone and the endometrium**

Successful implantation is the end result of complex molecular interactions between the hormonally primed uterus and an activated blastocyst. The failure to synchronize the component processes involved in these mechanisms results in a failure of implantation. Of the many aspects of the synchronization process, the role of steroid hormones is the best understood. The pre-ovulatory increase in the secretion of 17β-oestradiol, which promotes the proliferation and differentiation of uterine epithelial cells, is followed by the production of progesterone, which induces the proliferation and differentiation of stromal cells (Norwitz et al., 2001).

Progesterone acts on the endometrium by means of specific receptors, A and B, which are proteins located in the nucleus of endometrial cells. Progesterone receptor synthesis is controlled by oestrogens through oestrogen receptors during the proliferative phase. By inhibiting the synthesis of oestrogen receptors, progesterone leads to a fall of both oestrogen and progesterone receptors (reviewed in Bergeron, 2000). In line with this theory, experimental studies reported down regulation of progesterone receptors epithelial cell expression during the luteal phase of the menstrual cycle (Lessey et al., 1988; Tuckerman et al., 2004).
Progesterone has been named the ‘hormone of pregnancy’ as, in preparing the endometrium for blastocyst implantation and inducing endometrial development, it is crucial to establish and maintain the pregnancy. Studies in animals and humans demonstrated that progesterone regulates the migration and proliferation of immune and inflammatory cell populations in the endometrium (reviewed in Choi et al., 2000).

**Progesterone supplementation in recurrent miscarriage**

Recurrent miscarriage is a vexing clinical problem facing about 1% of couples attempting pregnancy. In up to 50% of cases, the exact underlying pathophysiological mechanisms remain unknown (Porter and Scott, 2005). There is evidence that recurrent miscarriage may be associated with retarded endometrial development in the peri-implantation period or luteal phase defect (Li et al., 2000). Two studies in which precisely timed endometrial specimens from women with recurrent miscarriage were examined reported incidences of LPD as high as 17.4% (Tulppala et al., 1991) and 28% (Li, 1998) respectively.

Interestingly, a growing body of evidence suggests that progesterone might play a significant role in establishing an adequate immune environment for the early stages of pregnancy (Piccinini et al., 1995; Choi et al., 2000; Kalinka and Szekeres-Bartho, 2005; Raghupathy et al., 2005).

Immunological recognition of pregnancy results in up-regulation of progesterone receptors on activated lymphocytes (Szekeres-Bartho et al., 1990). In the presence of progesterone, lymphocytes of pregnant women synthesize a 34-kDa protein known as progesterone-induced blocking factor (PIBF) (Szekeres-Bartho et al., 1985), which mediates both the immunomodulatory (Szekeres-Bartho et al., 1989) and antiabortive (Szekeres-Bartho et al., 1997a,b) properties of progesterone.

During pregnancy, the maternal immune system is modulated by progesterone via control of cytokine production (Szekeres-Bartho et al., 1997b). In normal pregnancies, there is a shift in the decidua from cellular immune response (Th1 cytokines) to humoral immunity (Th2 cytokines), which is highly controlled by PIBF (Szekeres-Bartho et al., 1996). Some authors postulated that significantly increased Th1 cytokine expression might represent the underlying phenomenon leading to reproductive failure (Ng et al., 2002). Further, the activation of peripheral blood mononuclear cells (PBMC) with trophoblast antigens confirmed that women with idiopathic recurrent spontaneous pregnancy loss have a Th1-type cytokine profile, characterized by production of interleukin (IL)-2, tumour necrosis factor (TNF) and interferon (IFN)γ (Raghupathy et al., 2000).

It was found that PIBF expression of maternal T-lymphocytes increases as a result of pregnancy and that the stimulus for PIBF induction occurs immediately after implantation (Check et al., 1996). Interestingly, Szekeres-Bartho et al. (1995) demonstrated that low percentage of PIBF-positive lymphocytes is inversely related to natural killer (NK) cell activity, preterm labour and pregnancy loss. The possible association between maternal Th1 dominance and recurrent miscarriage provides researchers with the challenge of trying to manipulate the Th1/Th2 cytokine balance to suppress the cell-mediated immunity. Progesterone has been proposed to act as an immunological suppressant blocking Th1 activity and inducing release of Th2 cytokines (IL-4 and IL-10).

Amongst the different types of progesterone, dydrogesterone has been the most widely used to support early pregnancy. Dydrogesterone is an orally active progestogen with high affinity for the progesterone receptors, being similar to endogenous progesterone in its pharmacological effects and molecular structure. Very recently, a prospective clinical trial (Raghupathy et al., 2005) investigated the effects of dydrogesterone therapy on Th1 and Th2 cytokine production in women with recurrent miscarriage. It was noted that progestogen down-regulates Th1 cytokines and stimulates Th2 cytokines, resulting in a shift towards Th2-type immunity. Dydrogesterone also induces PIBF production. In a randomized clinical trial, El-Zibdeh (2005) showed that therapeutic intervention with oral dydrogesterone significantly improves the chances of a successful pregnancy in women with recurrent miscarriage.

Human (Kalinka and Szekeres-Bartho, 2005) and animal (Joachim et al., 2003) studies suggested that inducing PIBF production could be the indirect mechanism by which dydrogesterone improves pregnancy outcome. Collectively, these data support the concept that progesterone might play an important role in the maintenance of pregnancy by inducing immune modulation at the endometrium level.

**Implantation failure in assisted reproduction**

Despite numerous developments in assisted reproduction, the implantation rate of replaced embryos remains low. It has been estimated that up to 85% of the embryos transferred to the uterus do not implant (Edwards, 1995). Even after repeated attempts at IVF or intracytoplasmic sperm injection (ICSI) and the replacement of morphologically normal embryos, about one-third of the patients do not become pregnant (de Mouzon et al., 1998; Engmann et al., 1999; Lurie et al., 2001; Check et al., 2002; Olivius et al., 2002; Schroder et al., 2004; Ubaldi et al., 2004). The repeated failure of implantation has been blamed on many factors and various approaches have been proposed to improve the success. These include immunological testing and treatment, allogenic lymphocyte therapy, intratubal transfer of zygotes and embryos, blastocyst transfer, sequential embryo transfer, assisted hatching, co-cultures, preimplantation genetic screening for aneuploidy and embryo donation (Urman et al., 2005a,b; Vaquero et al., 2006). Nevertheless, the inadequacy of the luteal phase remains an important cause of implantation failure in patients undergoing assisted reproduction.

**Luteal phase in assisted reproduction**

The adequacy of the luteal phase has been questioned since the early days of assisted reproduction, as it was assumed that the aspiration of the granulosa cells surrounding the oocyte could interfere with the production of progesterone, which is
necessary for successful implantation of the embryo. In 1984, Dlugi et al. studied the pattern of peri-ovulatory and luteal phase concentrations of serum oestradiol and progesterone in human menopausal gonadotrophin (HMG)-stimulated patients undergoing IVF and in controls. Despite the fact that peri-ovulatory serum oestradiol and mid-luteal progesterone concentrations were significantly higher in pregnant compared with non-pregnant patients, a decline in both plasma steroids during the mid-luteal phase was also observed in pregnant patients, thus suggesting some degree of corpus luteum deficiency (Dlugi et al., 1984). Similar results were reported by other authors who studied the luteal phase length and hormonal profiles in 77 patients undergoing IVF (Yovich et al., 1984). Gronow et al. (1985) studied 372 IVF patients treated with clomiphene citrate alone, clomiphene citrate and HMG, and HMG alone. They found that some non-conceptual cycles might have suffered early corpus luteal regression and suggested luteal phase support of patients treated with HMG. Subsequent studies reported LPD in many, but not all, IVF patients stimulated with HMG alone or a combination of HMG and clomiphene citrate (Huang et al., 1986; Lejeune et al., 1986; Zarutskie et al., 1987; Laitikainen et al., 1988; Van Steirteghem et al., 1988; Nylund et al., 1990).

Following the introduction of GnRH agonists in assisted reproduction, it became clear that the use of these compounds resulted in luteal phase insufficiency (Smits et al., 1987, 1992a). In a study by Smits et al. (1988), inadequate endometrial development was observed after ovarian stimulation with GnRH agonist (buserelin) and HMG and lack of luteal supplementation. The authors suggested that inadequate corpus luteum activity could be related to the prolonged blockage of pituitary gonadotrophic function following arrest of the GnRH agonist. Luteal phase insufficiency is more pronounced with GnRH agonist long protocols compared with short protocols (Devreker et al., 1996), and is present even after early cessation of GnRH agonist administration (Beckers et al., 2000). Furthermore, several studies (Albano et al., 1998; Diederich et al., 2001; Beckers et al., 2003; Kolibianakis et al., 2005) reported LPD in patients treated with GnRH antagonists, although the effect is milder than that of GnRH agonists (Lin et al., 1999).

Luteal phase insufficiency occurs regardless of the drug used to induce final oocyte maturation. In a recent study, the luteal phase was investigated in 39 IVF patients receiving GnRH antagonist regimens (Beckers et al., 2003). The final maturation of the follicles was triggered by recombinant human chorionic gonadotrophin (HCG) in 11 patients, recombinant LH in 13 patients and GnRH agonist (triptorelin) in 15 patients. Luteal phase insufficiency was found in all three groups of patients, although the luteal phase was less disturbed in the recombinant HCG group. Albano et al. (1999) reported that HCG administration in patients receiving GnRH antagonists results in the normalization of the luteal phase.

Data in the literature show that LPD in stimulated cycles is associated with advanced endometrial histological maturity (Forman et al., 1989; Macklon and Fauser, 2000; Tavaniotou et al., 2002; Saadat et al., 2004). In the study by Forman et al. (1989), nine out of 12 stimulated cycles showed advanced endometrial histological maturity compared with four natural control cycles. The authors concluded that the advanced endometrial maturation observed in stimulated IVF cycles is a consequence of the production of supraphysiologically concentrations of sex steroids by the corpus luteum, which cause profound modifications of endometrial receptor dynamics. Advanced endometrial histology has been observed in patients treated with GnRH agonists (Ubaldi et al., 1997) as well as GnRH antagonists (Kolibianakis et al., 2002; Papanikolaou et al., 2005).

Although the mechanism by which implantation is affected in stimulated cycles is not exactly known, a decrease in the concentration of cytoplasmic progesterone receptor, rather than a decrease in mid-luteal plasma progesterone concentrations, is likely to be involved (Forman et al., 1989; Molina et al., 1989). This in turn affects the chain of events leading to implantation (Simon et al., 2000). Forman et al. (1989) found a significant negative correlation between both pre-ovulatory oestradiol and day 16 progesterone and the concentration of cytosolic progesterone receptor (cPR), while advanced endometrial maturity tended to be associated with low concentrations of cPR. Further, natural cycles were characterized by low cytosolic oestradiol receptors (cER) and high cPR, whereas the concentration of both receptors was greatly reduced in stimulated cycles. Molina et al. (1985) measured luteal cytoplasmic oestradiol and progesterone receptor concentrations on day 22–25 endometrial samples obtained from seven IVF patients stimulated with clomiphene and HMG and from seven normally menstruating women. They noted that the endometrial concentration of cPR decreased in stimulated cycles, but cER remained unchanged.

Early studies reported unchanged progesterone concentrations in IVF patients treated with GnRHa/HMG regimens. Urbancsek et al. (1990) studied changes in serum oestradiol, LH and progesterone concentrations in 31 IVF patients stimulated with buserelin/HMG and 57 patients stimulated with HMG only and observed no significant differences in the luteal phase progesterone concentrations between the groups. Other authors (Hassiakos et al., 1990) investigated the oestradiol and progesterone concentrations, pregnancy and implantation rates in 565 IVF cycles stimulated with HMG alone and in 320 cycles stimulated with adjunctive treatment of a GnRH agonist (leuprolide). They found that the overall pattern of serum oestradiol and progesterone concentrations and the oestradiol/progesterone ratio were similar in both groups, though significantly higher absolute concentrations were demonstrated in leuprolide cycles. Balasch et al. (1991) assessed the luteal phase in 21 IVF patients who had no embryo transfer and found supraphysiologically plasma concentrations of oestradiol and progesterone in the mid-luteal phase of all women. Brzyski et al. (1991) examined the impact of pituitary suppression with GnRH agonists on the luteal phase in a population of IVF women who underwent identical stimulation regimens with and without leuprolide acetate. Serum progesterone concentrations measured during the luteal phase were significantly greater in the GnRHa cycles compared with the non-GnRHa cycles, but were similar when calculated per oocyte retrieved.
Progesterone to reduce implantation failure in assisted reproduction

The optimal time for starting progesterone administration for luteal support is still a matter of debate. In a randomized controlled trial (RCT), Sohn et al. (1999) found that starting intramuscular (i.m.) progesterone supplementation before oocyte retrieval negatively affects the implantation rate. More recently, another RCT showed no significant differences in the ongoing pregnancy rate when vaginal progesterone was started on the day of HCG administration, the day of oocyte retrieval or the day of embryo transfer (Moctar et al., 2006).

Numerous studies have reported the use of progesterone for luteal phase support in IVF cycles with various degrees of success (Yovich et al., 1984; Belaish-Allart et al., 1987; Kupferminc et al., 1990; Artini et al., 1995). In a recent Cochrane review (Tables 1–3), Daya and Gunby (2004) found that in IVF patients treated with HMG only, the odds of ongoing pregnancy was significantly increased, while the odds of clinical pregnancy and miscarriage did not reach statistical significance. In patients treated with GnRH agonists, the odds of ongoing pregnancy, clinical pregnancy and miscarriage did not show statistically significant differences. As yet, there has been no RCT evaluating the use of progesterone for luteal support in IVF patients treated with GnRH antagonists.

Progesterone versus HCG for luteal support in assisted reproduction

It has been suggested that HCG may offer a better alternative for luteal phase support compared with progesterone in IVF patients (Van Steirteghem et al., 1988; Claman et al., 1992; Golan et al., 1993; Araujo et al., 1994; Martinez et al., 2000). Conversely, Daya and Gunby (2004) found no significant difference in rates of clinical pregnancy, ongoing pregnancy or miscarriage between IVF patients who received progesterone and those who had HCG for luteal support (Tables 1–3). In HMG only regimen, the odds ratio for ongoing pregnancy and clinical pregnancy in HCG group versus progesterone group did not reach statistical significance. Similar findings were found in the GnRH agonist regimens. Of interest, the risk of ovarian hyperstimulation syndrome (OHSS) was about half when progesterone was used, compared with HCG (OR 0.46 (95% CI 0.26–0.81)), therefore making progesterone a more attractive option.

Progesterone and HCG for luteal support in assisted reproduction

A number of fertility physicians have recommended the combined administration of progesterone and HCG for luteal support in assisted reproduction (Herman et al., 1996; Moctar et al., 1996; Ludwig et al., 2001; Fujimoto et al., 2002). However, in their meta-analysis, Daya and Gunby (2004) found no statistically significant difference in rates of clinical pregnancy, ongoing pregnancy or miscarriage when progesterone plus HCG was compared with progesterone alone (Tables 1–3).

Further, the odds ratio of OHSS was more than 3-fold higher when HCG was added to the luteal phase support regimen [OR 3.06 (95% CI 1.59–5.86)] confirming that progesterone alone is a better strategy.

Progesterone and oestradiol for luteal support in assisted reproduction

Combining oestradiol and progesterone for luteal support in assisted reproduction patients has also been suggested (Smits, 1993; Lewin et al., 1994; Gleich et al., 2000; Farhi et al., 2000; Gorkemli et al., 2004). The Cochrane review by Daya and Gunby (2004) showed no significant difference in clinical pregnancy, ongoing pregnancy, miscarriage or live birth rates when progesterone combined with oestrogen was compared with progesterone alone for luteal support (Tables 1–3).

The addition of phytoestrogens to progesterone for luteal phase support has been recently investigated in an RCT. Unter et al. (2004a) studied 213 infertile patients undergoing IVF and embryo transfer and found that those who received progesterone (50 mg daily) plus phytoestrogens (1500 mg daily) had significantly higher implantation, clinical pregnancy and ongoing pregnancy/delivery rates compared with patients receiving progesterone and placebo. Larger studies are needed to further evaluate this interesting combination regimen before routine application to clinical practice.

Progesterone preparations, route of administration and teratogenicity

Regardless of the exact mechanism by which implantation is affected in hyperstimulated patients, various progestational compounds have been used for luteal support. These include natural progesterone and its derivatives as well as synthetic progestagens and their derivatives. Natural progesterone is quickly inactivated when taken orally, so synthetic derivatives (17α-hydroxyprogesterone derivatives and 19-nortestosterone derivatives) were developed to improve bioavailability (Posaci et al., 2000).

Micronization of natural progesterone has resulted in the development of oral progesterone preparations that can also be used vaginally (Miles et al., 1994). More recently, vaginal progesterone gels have been adopted for luteal support (Chantilis et al., 1999). Vaginal progesterone administration offers many advantages over oral administration, including rapid absorption, avoiding the first pass liver metabolism, providing higher bioavailability and diminishing the side effects, particularly nausea and drowsiness (Devroey et al., 1989; Bourgain et al., 1990; Smits et al., 1992b; Fanchin et al., 1997). Other routes of administration include the intranasal (Cicinelli et al., 1991), rectal (Chakmakjian and Zachariah, 1987) and sublingual (Stovall et al., 1996). Without doubt, the optimal route of administration and the optimal progesterone preparation used for luteal support in IVF have engendered controversy (Smits et al., 1992b; Artini et al., 1995; Pouly et al., 1996; Abate et al., 1997; Chantilis et al., 1999; Damario et al., 1999; Friedler et al., 1999; Costabile et al., 2001; Propst et
Table 1. Odds ratio (OD) and 95% confidence intervals of clinical pregnancy with various regimens used for luteal support in IVF patients (from Daya and Gunby, 2004).

<table>
<thead>
<tr>
<th>Comparison</th>
<th>OD (95% CI)</th>
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<tbody>
<tr>
<td>Progesterone versus placebo (HMG only regimen)</td>
<td>1.44 (0.97–2.14)</td>
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<tr>
<td>Progesterone versus placebo (GnRHa regimens)</td>
<td>1.25 (0.83–1.88)</td>
</tr>
<tr>
<td>HCG versus progesterone (HMG only regimen)</td>
<td>1.12 (0.64–1.96)</td>
</tr>
<tr>
<td>HCG versus progesterone (GnRHa regimens)</td>
<td>1.06 (0.82–1.36)</td>
</tr>
<tr>
<td>Progesterone + HCG versus progesterone only</td>
<td>1.10 (0.84–1.43)</td>
</tr>
<tr>
<td>Progesterone + oestradiol versus progesterone only</td>
<td>0.89 (0.43–1.84)</td>
</tr>
<tr>
<td>Intramuscular versus oral administration</td>
<td>2.28 (0.90–5.82)</td>
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<tr>
<td>Vaginal versus oral administration</td>
<td>1.51 (0.93–2.45)</td>
</tr>
<tr>
<td>Vaginal versus i.m. administration</td>
<td>0.82 (0.67–1.01)</td>
</tr>
<tr>
<td>Vaginal gel versus other vaginal administration</td>
<td>1.10 (0.67–1.82)</td>
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GnRHa = gonadotrophin-releasing hormone agonist; HCG = human chorionic gonadotrophin; HMG = human menopausal gonadotrophin.

Table 2. Odds ratio (OD) and 95% confidence intervals of ongoing pregnancy with various regimens used for luteal support in IVF patients (from Daya and Gunby, 2004).

<table>
<thead>
<tr>
<th>Comparison</th>
<th>OD (95% CI)</th>
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<tbody>
<tr>
<td>Progesterone versus placebo (HMG only regimen)</td>
<td>1.72 (1.09–2.72)*</td>
</tr>
<tr>
<td>Progesterone versus placebo (GnRHa regimens)</td>
<td>0.98 (0.57–1.67)</td>
</tr>
<tr>
<td>HCG versus progesterone (HMG only regimen)</td>
<td>1.08 (0.59–1.97)</td>
</tr>
<tr>
<td>HCG versus progesterone (GnRHa regimens)</td>
<td>0.90 (0.64–1.27)</td>
</tr>
<tr>
<td>Progesterone + HCG versus progesterone only</td>
<td>1.05 (0.69–1.60)</td>
</tr>
<tr>
<td>Progesterone + oestradiol versus progesterone only</td>
<td>0.89 (0.34–2.32)</td>
</tr>
<tr>
<td>Intramuscular versus oral administration</td>
<td>2.57 (0.99–6.70)</td>
</tr>
<tr>
<td>Vaginal versus oral administration</td>
<td>1.32 (0.79–2.19)</td>
</tr>
<tr>
<td>Vaginal versus i.m. administration</td>
<td>0.73 (0.56–0.96)*</td>
</tr>
<tr>
<td>Vaginal gel versus other vaginal administration</td>
<td>1.14 (0.62–2.10)</td>
</tr>
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GnRHa = gonadotrophin-releasing hormone agonist; HCG = human chorionic gonadotrophin; HMG = human menopausal gonadotrophin.

*Statistically significant.
Table 3. Odds ratio (OD) and 95% confidence intervals of miscarriage with various regimens used for luteal support in IVF patients (from Daya and Gunby, 2004).

<table>
<thead>
<tr>
<th>Comparison</th>
<th>OD (95% CI)</th>
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<tbody>
<tr>
<td>Progesterone versus placebo (HMG only regimen)</td>
<td>0.52 (0.23–1.18)</td>
</tr>
<tr>
<td>Progesterone versus placebo (GnRHa regimens)</td>
<td>1.33 (0.50–3.57)</td>
</tr>
<tr>
<td>HCG versus progesterone (HMG only regimen)</td>
<td>0.76 (0.23–2.47)</td>
</tr>
<tr>
<td>HCG versus progesterone (GnRHa regimens)</td>
<td>1.47 (0.69–3.14)</td>
</tr>
<tr>
<td>Progesterone + HCG versus progesterone only</td>
<td>1.70 (0.77–3.76)</td>
</tr>
<tr>
<td>Progesterone + oestradiol versus progesterone only</td>
<td>1.10 (0.18–6.76)</td>
</tr>
<tr>
<td>Intramuscular versus oral administration</td>
<td>0.40 (0.05–3.08)</td>
</tr>
<tr>
<td>Vaginal versus oral administration</td>
<td>1.11 (0.23–5.29)</td>
</tr>
<tr>
<td>Vaginal versus i.m. administration</td>
<td>0.81 (0.46–1.43)</td>
</tr>
<tr>
<td>Vaginal gel versus other vaginal administration</td>
<td>1.56 (0.35–7.01)</td>
</tr>
</tbody>
</table>

GnRHa = gonadotrophin-releasing hormone agonist; HCG = human chorionic gonadotrophin; HMG = human menopausal gonadotrophin.

Initially, progesterone was given by i.m. injection but the introduction of micronized progesterone preparation has allowed oral administration of the drug. Many researchers have compared both routes of administration. In their meta-analysis, Daya and Gunby (2004) found a 2-fold increase in the odds of both clinical and ongoing pregnancy with the IM route compared with the oral route, but this did not reach statistical significance. Similarly, when vaginal administration was compared with oral administration, no significant difference between the routes was observed in the rates of clinical or ongoing pregnancy. When vaginal administration was compared with IM administration, the 18% decrease in the odds of clinical pregnancy with vaginal progesterone was not statistically significant, while the ongoing pregnancy rate and the live birth rate were significantly diminished. Finally, comparing vaginal gel with the other types of vaginal administration, there was no statistically significant difference in the clinical pregnancy or ongoing pregnancy rates. These data are summarized in Tables 1–3.

Early studies have shown that progesterone administration during early pregnancy may be associated with an increased incidence of hypospadias. Some authors reported a 5-fold increased risk of hypospadias in male offspring conceived by IVF (Silver et al., 1999). Recently, in a population-based, multistate, case–control study, Carmichael et al. (2005) found that periconceptional progestin intake was associated with increased risk of hypospadias [OR 3.7 (95% CI 2.3–6.0)]. Therefore, both clinicians and patients should be aware of the potential teratogenic properties of maternal progesterone supplementation.

Conclusions

Luteal phase defect is an important cause of recurrent miscarriage and implantation failure after assisted reproduction. The hypothesis that an inadequately developed endometrium during the window of implantation may contribute to infertility is appropriate. Regardless of the mechanism of action, progesterone supplementation results in improving the chances of a successful pregnancy in both conditions. This is probably mediated by blocking Th1 activity and inducing release of Th2 cytokines, which favour embryo implantation. At least one RCT has shown the benefit of oral dydrogesterone in women with recurrent miscarriage.

Stimulation regimens used in assisted reproduction are invariably associated with some degree of luteal insufficiency. A recent meta-analysis of RCT shows that progesterone supplementation in women undergoing assisted reproduction resulted in a trend towards improved clinical and ongoing pregnancy rates but the odds ratios fall short of statistical significance, except for HMG-only regimens. The optimal progestational preparation and route remains a matter of debate, although IM administration of progesterone results in significantly higher ongoing pregnancy rates compared with vaginal administration. Patients have to be aware of the risk of hypospadias associated with maternal progestin intake.

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