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Female Fertility: What Every Urologist Must Understand

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Infertility is a disease of couples, and it affects 2.1 million married couples, many of whom seek fertility assistance [1]. Once thought to be a disease of women, it is clear that the cause of infertility in couples seeking advanced reproductive technology can be found in the male partner (18.5%), female partner (50%), both partners (18.4%), or neither partner (unexplained infertility, 12%). In 2006, 8% of married childless couples sought infertility services, and 15% of all women report having used some infertility service in the past. It behooves urologists who care for infertile men to be acquainted with female fertility. This article provides an overview of the causes for and treatment of female factor infertility.

Female reproductive physiology

In contrast to spermatogenesis, oogenesis begins in utero. By the fifth month of gestation, mitosis is complete and the peak number of oocytes is achieved. Barring any causes of ovarian failure, a pubertal girl has approximately 400,000 oocytes resting in primordial follicles containing a single layer of granulosa cells. During embryologic oogenesis, oocyte development arrests in the diplotene phase of the first meiotic division and does not resume meiosis until after ovulation. The process of ovulation requires that the follicle develop from a primordial follicle to a tertiary follicle over approximately three menstrual cycles. Only the final phase of folliculogenesis is hormone dependent. The process continues throughout postpubertal life even in the presence of cycle disruptors such as oral contraception or pregnancy. Follicles that reach the hormone-dependent stage at a time asynchronous with the menstrual cycle become atretic. Subsequently, only 300 to 400 follicles ovulate in a woman's lifetime.

The first day of the menstrual cycle is marked by the first day of bleeding. At this point, ovarian and pituitary hormone productions are at their nadir, and multiple morphologically identical antral follicles are present. Under the control of gonadotropin-releasing hormone from the hypothalamus, the anterior pituitary begins to release follicle-stimulating hormone (FSH) to begin hormone-dependent follicular growth. Cumulus expansion of the granulosa cells occurs as the oocytes compete to be the dominant follicle. Although selection of the dominant follicle is poorly understood, it is clear that the intrafollicular environment plays a significant role. The follicular fluid of the dominant follicle has significantly higher levels of estrogen than its competing cohort. There is also a greater concentration of luteinizing hormone (LH) receptor on the theca cells and FSH receptor on the granulosa cells. Several authors have theorized that the ability to recruit steroid receptors and produce estrogen is the major determinant of dominance [2]. Human animal data suggest that insulin-like growth factor (IGF) plays a major role in dominance selection; the *Igf* null mouse does not undergo hormone-dependent folliculogenesis [3,4]. In the bovine model, the dominant follicle has a low concentration of IGF binding proteins and an elevated concentration of pregnancy-associated plasma protein-A and insulin-like growth factor binding protein (IGFBP) proteolytic enzyme [5].

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A significant association between intrafollicular pregnancy-associated plasma protein-A and estradiol levels suggests that pregnancy-associated plasma protein-A plays a role in dominance selection in humans [6]. IGF is synergistic with FSH to augment estradiol production [7]. Data suggest that increase in intrafollicular-free IGF concentration is associated with dominance.

Clearly, the induction of estradiol is a key component of the follicular cycle. Initially, this estrogen has a negative feedback on the hypothalamus and pituitary. After follicular dominance is determined, however, estradiol exerts positive feedback that leads to the surge of LH. Again, the mechanisms involved in this surge are unclear. In addition to inducing ovulation, LH is required for the resumption of meiosis [8]. At the time of ovulation, the oocyte completes meiosis I and arrests in the metaphase of meiosis II. Meiosis is completed at the time of fertilization. The mechanisms involved in meiosis resumption are under intense study by researchers interested in in vitro maturation of follicles for preservation of fertility.

After ovulation, the ruptured follicle forms a corpus luteum and produces progesterone to support the endometrium for implantation. Implantation typically occurs 5 to 7 days after ovulation. The early trophoblast cells produce human chorionic gonadotropin to maintain the corpus luteum. In the absence of human chorionic gonadotropin, the corpus luteum involutes and the resultant decrease in estrogen and progesterone results in menses. Corpus luteal hormone production is critical to maintaining the pregnancy during the first 8 weeks of gestation; loss of corpus luteal progesterone production results in abortion.

Evaluation of the female partner

History

A focused history can evaluate for risk factors of infertility and guide further diagnostic modalities and treatment. Risk factors include advancing age, a history of pelvic infections (including salpingitis and appendicitis), severe pelvic pain or endometriosis, and irregular menstrual cycles. One also must ascertain the frequency of intercourse and the use of vaginal lubricants that may be sperm toxic. Of these risk factors, age is the strongest predictor of pregnancy either with or without advanced reproductive techniques. Fecundability begins to decline at age 30, sharply declines after age 35, and is less than 5% after age 40 (Fig. 1). Patients of advancing age are often counseled to begin with more aggressive treatment.

Menstrual history

Characterization of a woman's menstrual regularity may provide insight into the normalcy of ovulation. Normal menstrual cycles range from 24 to 25 days, and menses occurs for 3 to 7 days. In a woman younger than age 35, a history of regular menstrual cycles is highly correlated with the presence of ovulation. This association is strengthened when menses are accompanied by monthly moliminal symptoms, including breast tenderness, bloating, or mood changes. A long cycle is often associated with anovulation. In contrast, a short cycle may be associated with ovulation, with an inadequate follicular phase leading to poor endometrial development or luteal phase deficiency.

Assessment of ovulation

Several tools are available for the assessment of ovulation, including basal body temperature charts (Fig. 2), assessment of mid-luteal progesterone, endometrial biopsy, and urinary LH prediction kits. Each of these tests, with the exception of urinary LH, can only retrospectively inform the user of ovulation. Basal body temperature is effective because progesterone production from the corpus luteum raises core body temperature by approximately 0.6°F, which provides a "biphasic" pattern of temperature. Temperature should be taken every morning, however, which

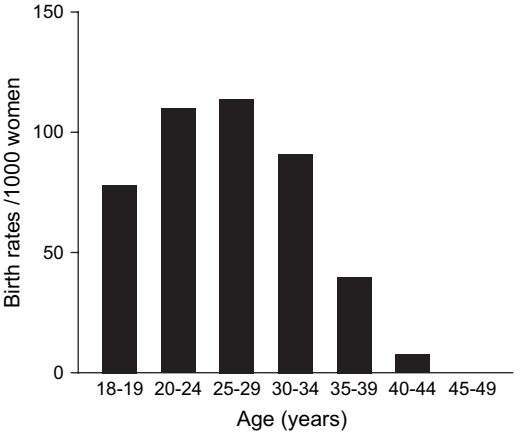

Fig. 1. Birth rates by maternal age, 2000. *Data from* National Vital Statistics Report. Available at: http://www.cdc.gov/nchs/data/statab/t001x07.pdf.

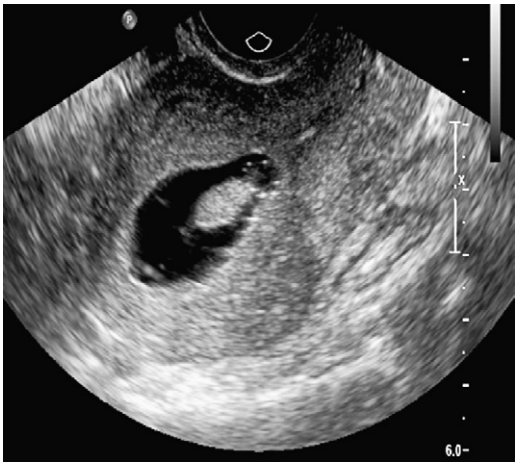

Fig. 2. Saline infusion sonogram shows an intrauterine polyp.

reminds the woman of infertility before starting her day. This reminder is cumbersome and may be emotionally taxing.

Measurement of mid-luteal progesterone is another retrospective method of determining ovulation. Clinicians often use this value to determine the adequacy of the luteal phase; however, data do not support a specific value that correlates with a luteal phase defect. Endometrial biopsy on cycle has long been considered the "gold standard" determinant of ovulation. After ovulation, the endometrium undergoes classic morphologic changes that correlate with the day of the menstrual cycle. It has been suggested the endometrial dysynchrony of two or more days, as determined by endometrial biopsy, is associated with luteal phase defect [9]. Significant inter- and intraobserver variability is present in the evaluation of endometrial biopsies, however [10,11]. The National Institutes of Health/National Institute of Child Health and Human Development–funded National Cooperative Reproductive Medicine Network recently completed a study on the use of endometrial biopsy. This multicentered trial concluded that the endometrial biopsy cannot discriminate between fertile and infertile women and should not be a routine part of the infertility evaluation [12].

Urinary LH testing uses an enzyme-linked immunoassay against the beta subunit of LH, which rises abruptly for approximately 18 hours before it peaks. Ovulation typically occurs approximately 36 hours after the onset of the surge. Because the hormone needs to be conjugated before it is excreted, urinary LH predicts ovulation approximately 24 hours in advance, which provides a prospective assay of ovulation that also can be used to time intercourse.

Ovarian reserve testing

Research has established that age is the best predictor of fertility potential. Ovarian reserve testing is a measure of "ovarian aging." Although studies have used these tests to predict the time to menopause, most studies have had in vitro fertilization (IVF) pregnancy as the outcome measure. Normative measures of ovarian reserve in a non-infertility population have not been established. Tests of ovarian reserve include the measurement of basal hormone levels, dynamic ovarian testing, and sonographic assessment of the ovaries.

Basal hormones that have been measured to estimate ovarian reserve include FSH, estradiol, inhibin, and anti-müllerian hormone. FSH is inversely proportional to the production of the follicular hormones estradiol and inhibin. It follows that as the cohort of developing follicles is reduced, the basal levels of FSH increase. Over the past 20 years, many studies have evaluated the predictive value of FSH, and the results of these studies are highly conflicting. Although most of these studies agree that the FSH level indicates the number of oocytes retrieved from an IVF cycle, several large studies have failed to demonstrate the test's ability to predict pregnancy, especially in a young patient population. Estradiol traditionally has been measured in conjunction with FSH to ensure that the FSH was drawn during the cycle nadir. Several studies have demonstrated that elevated estradiol level on day three may be independently predictive of poor stimulation, however [13,14]. Because neither of these hormones is highly predictive of pregnancy, investigators have tested the predictive value of additional hormones. Inhibin B, a hormone made by the granulosa cells, regulates FSH production by negative feedback. Studies suggest that inhibin B levels decrease earlier than changes in estradiol. Although results are conflicting, this hormone may be an earlier predictor of IVF response [15,16].

Dynamic tests of ovarian reserve involve challenging the ovaries with a fertility medication, such as clomiphene citrate or gonadotropin [17,18]. Although predictive of response to IVF, these tests have not been demonstrated to be superior to basal hormone levels in predicting pregnancy. Likewise, ultrasound ovarian assessments, including ovarian volume and antral follicle

counts, may predict response to medications. It is important to re-emphasize that although each test of ovarian reserve may help to counsel patients regarding their fertility potential, age remains the strongest predictor of pregnancy.

Pelvic infections and anatomic abnormalities

Pelvic infections, especially gonorrhea and chlamydia, may irreversibly damage the fallopian tubes. These infections are endemic in the United States and have an annual incidence of approximately 1 million new reported cases. Most of the cases are diagnosed in women between the ages of 15 and 24, with an annual incidence of approximately 1 in 35 women [19]. Unfortunately, because chlamydia is asymptomatic in 75% of cases, a large amount of chlamydial infections are not diagnosed. Up to 20% of women with undiagnosed chlamydia develop infertility. Although gonorrhea and chlamydia are the most common infectious causes of tubal damage, any significant pelvic inflammatory condition can result in adhesion formation that affects tubal integrity.

Severe dysmenorrhea may be associated with uterine anomaly, such as subserosal fibroids or outflow tract obstruction. It also may be associated with endometriosis or pelvic adhesions. This finding also leads to specific diagnostic evaluation.

Anatomic assessment

Assessment of the structural integrity of the reproductive tract is an essential component of the fertility evaluation. It can consist of radiologic imaging or surgical evaluation. The most common test for evaluating the uterine cavity and checking tubal patency is the hysterosalpingogram, which is performed by injecting radiopaque dye into the uterus and tubes under fluoroscopic visualization. Uterine abnormalities are outlined by the dye, and tubal obstruction is noted by the absence of free-spill into the peritoneal cavity. In addition to the diagnostic value of the hysterosalpingogram, the test may be therapeutic [20,21].

Intrauterine abnormalities also can be identified with saline-infusion sonography (Fig. 3). Because the uterus is a potential space, traditional ultrasonography is not sensitive enough to determine if a lesion is intracavitary. Saline-infusion sonography is performed by injecting saline into the uterus to provide a sonographic window within the endometrial cavity. The sensitivity and specificity of saline-infusion sonography have both been estimated to be 100% when surgery was used as a gold standard [22,23]. The advent of three-dimensional ultrasonography has improved the diagnostic capabilities of ultrasonography, and several publications have reviewed its technical aspects [24–26]. Briefly, the operator

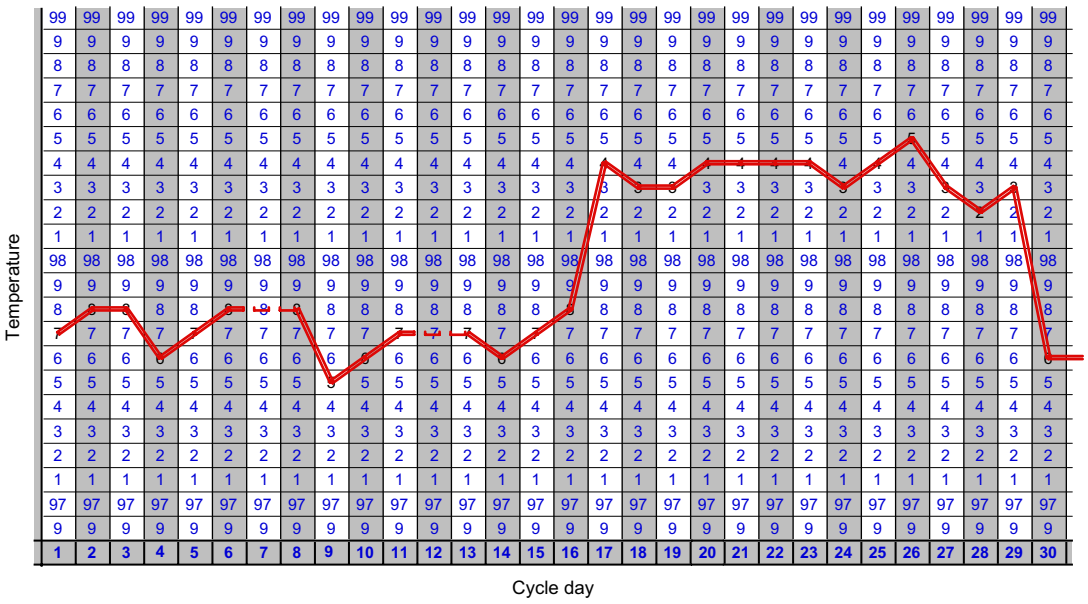

Fig. 3. Biphasic basal body temperature chart from an ovulatory patient.

selects a region of interest, and specialized probes scan through the region in multiple planes. Computer reconstruction assembles the images to create a structure that can be manipulated and viewed in multiple planes. This technique is highly accurate for diagnosing uterine anomalies and intrauterine pathologic conditions, such as septum and synechiae [27].

MRI is an excellent modality for viewing soft tissues, thereby surpassing the diagnostic ability of CT for imaging uterine abnormalities. It has been reported to have 100% specificity and 80% to 100% sensitivity for evaluating pelvic anomalies [28,29]. Because it images in multiple planes, MRI is an excellent preoperative assessment before reproductive gynecologic surgeries such as myomectomy and metroplasty.

Clearly, endoscopy can be used to visualize pelvic anatomy. Although radiologic imaging provides information about the pelvic structures, it provides little information about peritubal adhesions, pelvic infection, or endometriosis. The decision to perform laparoscopy as an initial diagnostic modality is based on the clinician's suspicion of pathologic condition. For example, in a patient with a history of cyclic pelvic pain that suggests endometriosis, laparoscopy may be the best initial evaluation.

Anatomic cervical abnormalities may result in abnormal cervical mucus production. Cervical mucus protects the sperm from the acidic milieu of the vagina and is critical for introducing sperm into the upper genital tract. Patients with extensive cervical surgery have lower fecundity. Some clinicians evaluate preovulatory cervical mucus with a postcoital test. Cervical mucus is evaluated for elasticity, ferning, and the presence of adequate numbers of motile sperm. The postcoital test has not been associated with improving pregnancy outcome, however.

Treatment strategies

Surgery

The use of tubal reconstruction has been highly debated in the infertility literature. In the early to mid-1990s, several authors reported that pregnancy rates after salpingostomy or fimbriplasty were equivalent to those associated with in vitro fertilization [30–32]. Variables that predict success include a patient's age, unilateral versus bilateral tubal disease, density of the adhesions, and thickness of the tubal wall. It should be noted that although pregnancy rates approximate 30%, ectopic pregnancy rates approximate 14% [33]. As the pregnancy rates from IVF continue to improve, the value of surgical intervention, with its increased surgical risk, has diminished. The exception to this may be tubal anastomosis after voluntary sterilization. Anastomosis is highly successful when at least 4 cm of tube are available for repair [34]. Although this allows the patient to have "natural conception," the cost and success are similar to those for IVF. For the couple who only wants to have one child or an additional child, anastomosis restores fertility and results in the need for birth control.

Laparoscopic surgery continues to be used for the treatment of endometriosis. Endometriosis can be found in 10% of all women and 30% to 40% of women with infertility [35,36]. Severe endometriosis can lead to peritoneal adhesions and a distortion of pelvic anatomy. Endometriosis also has been associated with an induction of peritoneal inflammation and oxidative stress, however, which has been associated with luteal dysfunction, poor embryonic development, and implantation failure [37]. Randomized clinical trials have been performed to determine if surgery improves outcome in patients with endometriosis [38,39]. Multiple meta-analyses have concluded that there is a benefit to surgical ablation of endometriosis, independent of the stage of disease [35,38]. The per-cycle pregnancy rate after IVF for women with endometriosis is higher than after surgery, however, despite the finding that endometriosis is associated lower peak E_2 concentration, a fewer number of oocytes retrieved, a lower fertilization rate, and a lower implantation rate [35]. Several studies have investigated the use of surgery to treat patients with endometriomas before infertility treatment. Although conflicting, most of these data do not demonstrate any clear advantage of surgical intervention [40–42].

Ovulation induction

Ovulation induction is the treatment of choice for patients with anovulation or unexplained infertility. For anovulatory patients, monofollicular development is the desired outcome. For unexplained infertility, superovulation is desired to increase the probability of conception. Medications used include selective estrogen receptor modulators, aromatase inhibitors, and gonadotropins, each of which has distinct advantages and disadvantages.

Clomiphene citrate is the most commonly used medication for the treatment of infertility. A selective estrogen receptor modulator, it is a competitive antagonist of estradiol at the level of the cytoplasmic nuclear receptor complex. The drug binds to estrogen receptors in the hypothalamic arcuate nucleus, disrupts the negative feedback, and augments gonadotropin-releasing hormone production. Endogenous production of FSH is augmented and hyperstimulation is achieved [43]. The main side effects of clomiphene therapy are related to the brain's decreased perception of estrogen. Patients often complain of hot flashes, headaches, and visual changes. Clomiphene citrate can compete with estrogen at receptor sites outside the brain, including the uterus and cervix. Lowering the effect of estrogen in the uterus can result in poor endometrial development and low implantation rates [44,45]. Estrogenized cervical mucus is necessary to provide an environment to support sperm survival and transport [46]. Clomiphene citrate has been associated with decreases in cervical mucus score that result in poor sperm-mucus interactions [47,48]. Some clinicians advocate performing postcoital tests in patients undergoing clomiphene citrate induction cycles or routinely performing intrauterine inseminations.

The aromatase inhibitors—letrozole and anastrozole—recently were reported to be beneficial for ovulation induction [49]. These drugs are competitive reversible inhibitors of testosterone aromatization and decrease circulating estrogen by more than 97%. Similar to clomiphene, the reduction in estrogen affects the hypothalamic feedback and induces greater levels of FSH. Because there is no suppression of the estrogen receptor, researchers have postulated that aromatase inhibitors would not negatively affect the uterus or cervical mucus, and several randomized controlled trials have demonstrated improved endometrial development with the use of letrozole [50,51]. Recent concern has been raised about the association of aromatase inhibitors with birth defects. This association seems unlikely because the medications are not present during the time of organogenesis because of their short half-lives (approximately 45 hours) [52]. A recent multicenter trial did not report any increased risk of birth defects in a cohort of 514 children born after letrozole stimulation [53].

The mainstay of ovulation induction, especially for assisted reproductive technologies, is exogenous gonadotropins. Gonadotropins were first purified from pregnant mares' urine more than 70 years ago. Although effective, the medication was highly antigenic and could only be used for limited cycles. Human menopausal gonadotropin became clinically available in the 1960s. Initially, human menopausal gonadotropin contained a 1:1 mixture of FSH:LH [54], and these initial preparations contained a large amount (> 90%) of protein impurities [55]. As purification techniques improved, the amount of LH in the preparations decreased. Currently, most FSH is pure FSH because it is produced in Chinese hamster ovary cells that contain genes inserted with recombinant technology [54]. Debate has arisen regarding the requirement of LH supplementation for folliculogenesis. During ovulation induction for patients without hypothalamic amenorrhea, a significant need for LH is unlikely [56]. Although most patients respond adequately to FSH alone after pituitary down-regulation for in vitro fertilization, there is likely to be a subset of patients who would benefit from LH supplementation. Additional research is necessary to identify this subgroup, however.

Typical starting doses of gonadotropin range between 75 IU and 225 IU, depending on a patient's age, diagnosis, and prior stimulation history. Patients should be monitored frequently with ultrasound and estradiol levels to assess follicular development and maturity, and the dose of gonadotropin should be adjusted to avoid overstimulation. When two to four follicles are approximately 18 mm in mean diameter, ovulation can be induced with human chorionic gonadotropin.

Ovulation induction and ovarian cancer risk

It has been hypothesized that use of fertility medications may increase a patient's risk for ovarian cancer. One of the first studies to make this association was by Whittemore and colleagues in 1993 [57]. This study was an analysis of data collected from 12 case-controlled studies of ovarian cancers diagnosed over a 30-year period. They reported a 2.7-fold increase in ovarian cancer in women who had taken fertility drugs and conceived and a 27-fold increase in ovarian cancer in women who took drugs and remained nulligravid. This study did not identify a precise "fertility drug" and included numerous drugs used to treat infertility. Their conclusion was met with numerous editorials that challenged the validity of their results.

Cohen and colleagues [58] concluded that the association between ovarian cancer and fertility drugs was caused by a confounding effect of gravidity; once gravidity was controlled, the association between drugs and cancer was not significant. The first major cohort study was published by Rossing and colleagues [59], who detected 11 ovarian tumors, of which 4 were invasive epithelial cancers, in 3837 women and reported a significant association with fertility drug use. The strongest association was in users of clomiphene citrate after 12 consecutive cycles (11-fold increase). Since that time, numerous epidemiologic studies have been published arguing for [60] and against [61,62] this association. The bulk of this evidence does not support the association between ovarian cancer and fertility drugs. The American Society of Reproductive Medicine concluded that "Although initial reports suggested that women who use fertility drugs have an increased risk for ovarian cancer, numerous recent studies support the conclusion that fertility drugs are not linked to ovarian cancer. Nevertheless, there is still uncertainty whether a risk exists and research continues to address this question" [63].

In vitro fertilization with or without intracytoplasmic sperm injection (ICSI)

Some researchers believe that in vitro fertilization is the most successful treatment for infertility, regardless of diagnosis. In 2004, 411 fertility clinics in the United States performed 127,977 IVF cycles that resulted in the births of 49,458 infants [64]. This statistic represents more than approximately 1.2% of all births in the United States. For women younger than age 35, the live birth rate/transfer is 43%, which is considerably higher than the live birth rate of approximately 30%/transfer in 1995. The improved pregnancy rates are largely attributable to improvements in embryo handling in the laboratory and at the time of transfer. In the laboratory, culture media have been improved by reducing the amount of glucose [65], reducing oxidative stress, and supplementing the media with amino acids [66] and growth factors [67]. Reports also suggest improved embryo development by culturing embryos in physiologic oxygen tension [68]. Atraumatic transfer catheters and ultrasound-guided transfer have contributed to improved pregnancy rates [69]. These improvements are coupled with better abilities to select the best embryos for transfer at the cleaved and blastocyst stages [68,70]. A more detailed description of ICSI can be found in the article by Alukal and Lamb elsewhere in this issue.

Summary

Treatments for infertility have been improving dramatically, and an increasing number of couples are seeking treatment. This is likely to translate to an increase in the number of infertility visits to urologists. Because abnormalities are frequently noted in both partners and treatment of the female partner can affect the treatment of the male partner, it is critical that urologists understand the basics of female infertility. This understanding also facilitates improved communication between the treating urologist and reproductive endocrinologist.

References

[1] Chandra A, Martinez GM, Mosher WD, et al. Fertility, family planning, and reproductive health of U.S. women: data from the 2002 National Survey of Family Growth. National Center for Health Statistics. Vital Health Stat 2005;23(25):2.

[2] Hillier SG, van den Boogaard AM, Reichert LE Jr, et al. Intraovarian sex steroid hormone interactions and the regulation of follicular maturation: aromatization of androgens by human granulosa cells in vitro. J Clin Endocrinol Metab 1980;50(4):640–7.

[3] Baker J, Hardy MP, Zhou J, et al. Effects of an Igf1 gene null mutation on mouse reproduction. Mol Endocrinol 1996;10(7):903–18.

[4] Echternkamp SE, Howard HJ, Roberts AJ, et al. Relationships among concentrations of steroids, insulin-like growth factor-I, and insulin-like growth factor binding proteins in ovarian follicular fluid of beef cattle. Biol Reprod 1994;51(5):971–81.

[5] Mihm M, Austin EJ, Good TE, et al. Identification of potential intrafollicular factors involved in selection of dominant follicles in heifers. Biol Reprod 2000;63(3):811–9.

[6] Conover CA, Faessen GF, Ilg KE, et al. Pregnancy-associated plasma protein-a is the insulin-like growth factor binding protein-4 protease secreted by human ovarian granulosa cells and is a marker of dominant follicle selection and the corpus luteum. Endocrinology 2001;142(5):2155–8.

[7] Adashi EY. The IGF family and folliculogenesis. J Reprod Immunol 1998;39(1–2):13–9.

[8] Conti M, Andersen CB, Richard FJ, et al. Role of cyclic nucleotide phosphodiesterases in resumption of meiosis. Mol Cell Endocrinol 1998;145(1–2):9–14.

[9] Wentz AC. Endometrial biopsy in the evaluation of infertility. Fertil Steril 1980;33(2):121–4.

[10] Myers ER, Silva S, Barnhart K, et al. Interobserver and intraobserver variability in the histological dating of the endometrium in fertile and infertile women. Fertil Steril 2004;82(5):1278–82.
[11] Scott RT, Snyder RR, Bagnall JW, et al. Evaluation of the impact of intraobserver variability on endometrial dating and the diagnosis of luteal phase defects. Fertil Steril 1993;60(4):652–7.
[12] Coutifaris C, Myers ER, Guzick DS, et al. Histological dating of timed endometrial biopsy tissue is not related to fertility status. Fertil Steril 2004;82(5): 1264–72.
[13] Licciardi FL, Liu HC, Rosenwaks Z. Day 3 estradiol serum concentrations as prognosticators of ovarian stimulation response and pregnancy outcome in patients undergoing in vitro fertilization. Fertil Steril 1995;64(5):991–4.
[14] Smotrich DB, Widra EA, Gindoff PR, et al. Prognostic value of day 3 estradiol on in vitro fertilization outcome. Fertil Steril 1995;64(6):1136–40.
[15] Seifer DB, Lambert-Messerlian G, Hogan JW, et al. Day 3 serum inhibin-B is predictive of assisted reproductive technologies outcome. Fertil Steril 1997; 67(1):110–4.
[16] Hall JE, Welt CK, Cramer DW. Inhibin A and inhibin B reflect ovarian function in assisted reproduction but are less useful at predicting outcome. Hum Reprod 1999;14(2):409–15.
[17] Hofmann GE, Sosnowski J, Scott RT, et al. Efficacy of selection criteria for ovarian reserve screening using the clomiphene citrate challenge test in a tertiary fertility center population. Fertil Steril 1996;66(1): 49–53.
[18] Winslow KL, Toner JP, Brzyski RG, et al. The gonadotropin-releasing hormone agonist stimulation test: a sensitive predictor of performance in the flare-up in vitro fertilization cycle. Fertil Steril 1991;56(4):711–7.
[19] Centers for Disease Control and Prevention. Sexually Transmitted Disease Surveillance 2005 Supplement, Chlamydia Prevalence Monitoring Project Annual Report 2005. Atlanta, GA: U.S. Department of Health and Human Services, Centers for Disease Control and Prevention, December 2006.
[20] Al-Fadhli R, Sylvestre C, Buckett W, et al. A randomized study of laparoscopic chromopertubation with lipiodol versus saline in infertile women. Fertil Steril 2006;85(2):505–7.
[21] Luttjeboer F, Harada T, Hughes E, et al. Tubal flushing for subfertility. Cochrane Database Syst Rev 2007;(3):CD003718.
[22] Keltz MD, Olive DL, Kim AH, et al. Sonohysterography for screening in recurrent pregnancy loss. Fertil Steril 1997;67(4):670–4.
[23] Sylvestre C, Child TJ, Tulandi T, et al. A prospective study to evaluate the efficacy of two- and three-dimensional sonohysterography in women with intrauterine lesions. Fertil Steril 2003;79(5): 1222–5.
[24] Bega G, Lev-Toaff AS, O'Kane P, et al. Three-dimensional ultrasonography in gynecology: technical aspects and clinical applications. J Ultrasound Med 2003;22(11):1249–69.
[25] Pairleitner H, Steiner H, Hasenoehrl G, et al. Three-dimensional power Doppler sonography: imaging and quantifying blood flow and vascularization. Ultrasound Obstet Gynecol 1999;14(2):139–43.
[26] Yaman C, Jesacher K, Polz W. Accuracy of three-dimensional transvaginal ultrasound in uterus volume measurements: comparison with two-dimensional ultrasound. Ultrasound Med Biol 2003;29(12): 1681–4.
[27] Makris N, Skartados N, Kalmantis K, et al. Evaluation of abnormal uterine bleeding by transvaginal 3-D hysterosonography and diagnostic hysteroscopy. Eur J Gynaecol Oncol 2007;28(1):39–42.
[28] Pellerito JS, McCarthy SM, Doyle MB, et al. Diagnosis of uterine anomalies: relative accuracy of MR imaging, endovaginal sonography, and hysterosalpingography. Radiology 1992;183(3):795–800.
[29] Zurawin RK, Dietrich JE, Heard MJ, et al. Didelphic uterus and obstructed hemivagina with renal agenesis: case report and review of the literature. J Pediatr Adolesc Gynecol 2004;17(2):137–41.
[30] Dlugi AM, Reddy S, Saleh WA, et al. Pregnancy rates after operative endoscopic treatment of total (neosalpingostomy) or near total (salpingostomy) distal tubal occlusion. Fertil Steril 1994;62(5): 913–20.
[31] Schlaff WD, Hassiakos DK, Damewood MD, et al. Neosalpingostomy for distal tubal obstruction: prognostic factors and impact of surgical technique. Fertil Steril 1990;54(6):984–90.
[32] Vasquez G, Boeckx W, Brosens I. Prospective study of tubal mucosal lesions and fertility in hydrosalpinges. Hum Reprod 1995;10(5):1075–8.
[33] Taylor RC, Berkowitz J, McComb PF. Role of laparoscopic salpingostomy in the treatment of hydrosalpinx. Fertil Steril 2001;75(3):594–600.
[34] Van Voorhis BJ. Comparison of tubal ligation reversal procedures. Clin Obstet Gynecol 2000;43(3): 641–9.
[35] Barnhart K, Dunsmoor-Su R, Coutifaris C. Effect of endometriosis on in vitro fertilization. Fertil Steril 2002;77(6):1148–55.
[36] Lessey BA, Castelbaum AJ, Sawin SW, et al. Aberrant integrin expression in the endometrium of women with endometriosis. J Clin Endocrinol Metab 1994;79(2):643–9.
[37] Kennedy S, Bergqvist A, Chapron C, et al. ESHRE guideline for the diagnosis and treatment of endometriosis. Hum Reprod 2005;20(10):2698–704.
[38] Jacobson TZ, Barlow DH, Koninckx PR, et al. Laparoscopic surgery for subfertility associated with endometriosis. Cochrane Database Syst Rev 2002;4:CD001398.
[39] Parazzini F. Ablation of lesions or no treatment in minimal-mild endometriosis in infertile women: a

randomized trial. Gruppo Italiano per lo Studio dell'Endometriosi. Hum Reprod 1999;14(5):1332–4.
[40] Al-Azemi M, Bernal AL, Steele J, et al. Ovarian response to repeated controlled stimulation in in-vitro fertilization cycles in patients with ovarian endometriosis. Hum Reprod 2000;15(1):72–5.
[41] Marconi G, Vilela M, Quintana R, et al. Laparoscopic ovarian cystectomy of endometriomas does not affect the ovarian response to gonadotropin stimulation. Fertil Steril 2002;78(4):876–8.
[42] Pabuccu R, Onalan G, Goktolga U, et al. Aspiration of ovarian endometriomas before intracytoplasmic sperm injection. Fertil Steril 2004;82(3):705–11.
[43] Dickey RP, Holtkamp DE. Development, pharmacology and clinical experience with clomiphene citrate. Hum Reprod Update 1996;2(6):483–506.
[44] Haritha S, Rajagopalan G. Follicular growth, endometrial thickness, and serum estradiol levels in spontaneous and clomiphene citrate-induced cycles. Int J Gynaecol Obstet 2003;81(3):287–92.
[45] Yagel S, Ben-Chetrit A, Anteby E, et al. The effect of ethinyl estradiol on endometrial thickness and uterine volume during ovulation induction by clomiphene citrate. Fertil Steril 1992;57(1):33–6.
[46] Moghissi KS, Syner FN, Evans TN. A composite picture of the menstrual cycle. Am J Obstet Gynecol 1972;114(3):405–18.
[47] Marchini M, Dorta M, Bombelli F, et al. Effects of clomiphene citrate on cervical mucus: analysis of some influencing factors. Int J Fertil 1989;34(2): 154–9.
[48] Randall JM, Templeton A. Cervical mucus score and in vitro sperm mucus interaction in spontaneous and clomiphene citrate cycles. Fertil Steril 1991; 56(3):465–8.
[49] Mitwally MFM, Casper RF. Aromatase inhibition: a novel method of ovulation induction in women with polycystic ovarian syndrome. Reprod Technol 2000;10:244–7.
[50] Atay V, Cam C, Muhcu M, et al. Comparison of letrozole and clomiphene citrate in women with polycystic ovaries undergoing ovarian stimulation. J Int Med Res 2006;34(1):73–6.
[51] Begum MR, Ferdous J, Begum A, et al. Comparison of efficacy of aromatase inhibitor and clomiphene citrate in induction of ovulation in polycystic ovarian syndrome. Fertil Steril 2008; [epub ahead of print].
[52] Tiboni GM. Aromatase inhibitors and teratogenesis. Fertil Steril 2004;81:1158–9.
[53] Tulandi T, Martin J, Al-Fadhli R, et al. Congenital malformations among 911 newborns conceived after infertility treatment with letrozole or clomiphene citrate. Fertil Steril 2006;85(6):1761–5.
[54] Ludwig M, Westergaard LG, Diedrich K, et al. Developments in drugs for ovarian stimulation. Best Pract Res Clin Obstet Gynaecol 2003;17(2): 231–47.
[55] Giudice E, Crisci C, Eshkol A, et al. Composition of commercial gonadotrophin preparations extracted from human post-menopausal urine: characterization of non-gonadotrophin proteins. Hum Reprod 1994;9(12):2291–9.
[56] Homburg R. Ovulation induction. Expert Opin Pharmacother 2003;4(11):1995–2004.
[57] Whittemore AS, Harris R, Itnyre J, et al. Characteristics relating to ovarian cancer risk: collaborative analysis of 12 US case-control studies. I. Methods. Collaborative Ovarian Cancer Group. Am J Epidemiol 1992;136(10):1175–83.
[58] Cohen J, Forman R, Harlap S, et al. IFFS expert group report on the Whittemore study related to the risk of ovarian cancer associated with the use of infertility agents. Hum Reprod 1993;8(7):996–9.
[59] Rossing MA, Daling JR, Weiss NS, et al. Ovarian tumors in a cohort of infertile women. N Engl J Med 1994;331(12):771–6.
[60] Kashyap S, Moher D, Fung MF, et al. Assisted reproductive technology and the incidence of ovarian cancer: a meta-analysis. Obstet Gynecol 2004; 103(4):785–94.
[61] Brinton LA, Lamb EJ, Moghissi KS, et al. Ovarian cancer risk after the use of ovulation-stimulating drugs. Obstet Gynecol 2004;103(6):1194–203.
[62] Venn A, Healy D, McLachlan R. Cancer risks associated with the diagnosis of infertility. Best Pract Res Clin Obstet Gynaecol 2003;17(2):343–67.
[63] ASRM. Assisted reproductive technologies: a guide for patients. Burmingham (AL): American Society of Reproductive Medicine; 2003. p. 13.
[64] Wright VC, Chang J, Jeng G, et al. Assisted reproductive technology surveillance-United States, 2004. MMWR Surveill Summary 2007;56(26):1–22.
[65] Conaghan J, Handyside AH, Winston RM, et al. Effects of pyruvate and glucose on the development of human preimplantation embryos in vitro. J Reprodu Fertil 1993;99(1):87–95.
[66] Devreker F, Winston RM, Hardy K. Glutamine improves human preimplantation development in vitro. Fertil Steril 1998;69(2):293–9.
[67] Spanos S, Becker DL, Winston RM, et al. Anti-apoptotic action of insulin-like growth factor-I during human preimplantation embryo development. Biol Reprod 2000;63(5):1413–20.
[68] Gardner D, Lane M. Embryo culture. In: Gardner D, Weissman A, Howles C, et al, editors. Textbook of assisted reproductive techniques: laboratory and clinical perspectives. London: Martin Dunitz Press; 2001. p. 203–22.
[69] Buckett WM. A review and meta-analysis of prospective trials comparing different catheters used for embryo transfer. Fertil Steril 2006;85(3):728–34.
[70] Dennis SJ, Thomas MA, Williams DB, et al. Embryo morphology score on day 3 is predictive of implantation and live birth rates. J Assist Reprod Genet 2006;23(4):171–5.