Approximately 15% of reproductive-age couples are infertile. While assisted reproductive technology (ART) offers these couples their best chance of having a family, ART itself has limitations. Two out of every three in vitro fertilization (IVF) cycles do not result in a pregnancy, and over 80% of embryos generated by the procedure fail to implant in a woman's uterus. It is not surprising then that many infertility centers attain relatively high success rates following IVF through the simultaneous transfer of multiple embryos. Consequently, more than 30% of IVF pregnancies are twins or higher-order multiple gestations, and 51% of all IVF neonates are the products of multiple gestations. Today, decreasing multiple gestations while maintaining or improving overall pregnancy rates remains the most important goal in the treatment of infertility.

Most clinicians understand the urgency of meeting this goal. Among women who carry more than one fetus, there is a two- to four-fold greater risk of pregnancy-induced hypertension and postpartum hemorrhage and a greater likelihood of preterm delivery, which in turn threatens an infant’s survival and increases the chances of lifelong disabilities such as cerebral palsy.

A key issue surrounding multiple gestations following IVF is the inability to precisely and non-invasively determine the viability of individual embryos. Shortly after the first IVF-generated birth occurred, researchers saw an association between an embryo's cleavage rate and morphologic characteristics and the ability of the embryo to implant and produce a live birth. This association eventually gave way to viability grading systems. While these grading systems, in combination with an improved culture environment, have increased implantation and pregnancy rates, they are far from ideal. They are still not precise enough to compel most patients and clinicians to reduce the number of embryos transferred to a point where twins are uncommon and higher-order multiple gestations are rare or eliminated entirely. In fact, even using the best morphologic and cleavage data, only about half of all IVF-generated embryos (49%) actually implant on day 3.

(CONTINUED ON PAGE 6)
Greetings from Charles J. Lockwood, MD, MHCM, The Anita O’Keeffe Young Professor of Women’s Health and Chair of Obstetrics, Gynecology and Reproductive Sciences at Yale University School of Medicine, and Chief of Ob/Gyn at Yale-New Haven Hospital.

Once again it is my distinct honor and privilege to tell you about the truly fascinating research projects ongoing in the Yale Department of Obstetrics, Gynecology and Reproductive Sciences. We were recently ranked fourth in the nation by the US News and World Report Survey for academic departments of women's health. We also reach over $11 million per year in total NIH funding.

We start with an article by Dr. Emre Seli on how measuring an embryo’s metabolism helps detect viable embryos. Emre uses near-infrared spectroscopic examination of IVF culture media to detect metabolomic patterns that strongly correlate with viable, usually euploid, embryos. This approach has already successfully predicted the implantation potential of embryos in a large clinical trial. We believe this rapid, non-invasive, relatively inexpensive test could decrease multiple gestations and improve IVF pregnancy rates in the very near future.

Equally exciting is the work of Dr. Hugh Taylor, who has been discovering and cataloging the many applications of stem cells derived from the endometrium. The Taylor lab has transformed these cells into insulin-producing pancreatic beta-cells and most recently into dopamine-producing neurons. Astonishingly, when these cells are transplanted into the brain of an animal with Parkinson’s disease, they have remarkable therapeutic effects.

Another Yale REI physician, Dr. Beth Rackow, tells about her research to determine how fibroids affect fertility. She has discovered that HOXA10 mRNA expression was significantly lower in the endometrium of women with submucosal myomas. Since HOXA10 expression is crucial to normal implantation, this finding helps explain why poor reproductive outcomes are often seen in such women, and also offers new strategies for restoring fertility.

Finally, my old colleague from Contemporary Ob/Gyn and our newsletter’s new executive editor, Paul Cerrato, and I discuss practical strategies for managing the obese gravida. I hope you enjoy reading about these topics as much as we enjoy talking about them.
Evidence suggesting that stem cells may have therapeutic value in Parkinson’s disease (PD) continues to mount. Investigators have shown that mesenchymal stem cells (MSCs), administered either intravenously or intracranially, can migrate to areas of the brain where damaged neurons are located.1 Others have taken this a step further, finding that transplanting human neural stem cells into the brains of monkeys with severe neurotoxin-induced PD produces significant improvements in “activities of daily living,” including their ability to sit, walk and feed themselves.2 And in a recent open-label pilot study, PD patients who had autologous bone-marrow-derived MSCs transplanted into the sublateral ventricular zone experienced subjective improvement in facial expression, gait and freezing episodes and no serious adverse reactions.3

While none of these studies indicate that stem cell therapy is ready for clinical use, they have opened the door for other researchers to look more closely at the possible mechanism of action of stem cells and explore other potentially useful treatments. At Yale, one of the most exciting avenues of research is adult human endometrium-derived stem cells (HEDSC).

The regenerative potential of endometrial cells
HEDSCs are a type of mesenchymal stem cell that takes advantage of the fact that the endometrium has potent regenerative powers, as evidenced by its ability to regrow with every menstrual cycle. Researchers have shown, for instance, that endometrial stem cells can be transformed into cartilage, bone, fat and muscle. In our laboratory, we recently demonstrated that they can also be transformed into dopamine-producing neurons. And when transplanted into the brain of an animal that has been given an experimental version of PD, we discovered that the cells have a therapeutic effect.4

To reach that conclusion, we initially collected endometrial tissue by curettage from nine reproductive-age women who were having gynecological surgery for benign conditions. These stromal cells were cultured and then subjected to a dopamnergic differentiation procedure. The resulting dopamine-producing cells demonstrated characteristic neuron morphology, enzyme activity and electrophysiologic properties. The cells were then transplanted into the striatum of both immunocompromised and immunocompetent mice that had been exposed to MPTP, a potent neurotoxin that destroys dopaminergic cells in animals and humans.

We were then able to confirm that the endometrial-derived stem cells migrated to the substantia nigra, the site where dopamine-producing brain cells die during PD. And further testing revealed that dopamine concentration went up in these transplanted animals.

If clinical trials support the safety and efficacy of this approach, clinicians would have a readily available therapeutic agent that does not require the controversial use of embryo-derived stem cells. The fact that the cells were not rejected by either immunocompromised or immunocompetent mice suggests that the therapy could be administered using either an autologous or allogenic protocol.

(CONTINUED ON PAGE 7)
Any clinician who has ever struggled to find the cause of a woman’s infertility is well aware of the uncertainty about the link between uterine myomas and infertility. Over the years, investigators have theorized that myomas may impair fertility by occluding the interstitial segment of the fallopian tube; by causing abnormal uterine contractions that interfere with ovum or sperm transport; or by interfering with endometrial blood flow, which in turn can cause focal endometrial attenuation or ulceration. Others have postulated that myomas may even act as foreign bodies, precipitating an inflammatory reaction that interferes with the ability of the embryo to successfully implant.1 Although these proposed mechanisms of action have merit, none examine the molecular association between myomas and infertility. In fact, histological evaluation of the endometrium has been unsuccessful in differentiating fertile from infertile women.

To address this issue, Yale researchers have begun to look at established molecular markers of endometrial receptivity and have recently identified differences between women with submucosal uterine myomas and those with normal uteri —differences that likely affect a woman’s reproductive potential.2

A closer look at the research

Our research team conducted a case-control study that examined endometrial tissue from 30 women: 14 had submucosal myomas, nine had intramural myomas with no uterine cavity distortion, and seven had no myomas. The women’s ages ranged from 31 to 48 years. None had any disorder that has been shown to affect endometrial receptivity such as PCOS, endometriosis or hydrosalpinges. In each woman, the endometrium was evaluated for levels of several established markers of endometrial receptivity; these markers have been well studied in mouse models and on human endometria, and have been shown to be necessary for implantation. Among the markers we measured was HOXA10, a transcription factor involved in embryonic uterine development and adult endometrial development during each menstrual cycle. The analysis also measured expression of HOXA11, a protein that likely plays a role in endometrial development and implantation; BTEB1 (basic transcriptional element binding protein), which researchers believe may be involved in endometrial development and blastocyst implantation.

The analysis found that endometrial HOXA10 mRNA expression was significantly lower in the endometrium overlying the submucosal myoma; but more importantly, we also found lower HOXA10 expression in the endometrium remote from the submucosal myoma; in contrast, women without myomas and those with intramural myomas had comparatively higher levels of endometrial HOXA10 mRNA expression. Similar findings were seen with endometrial HOXA11 mRNA expression. Furthermore, immunohistochemistry localized the decrease in endometrial HOXA10 expression to the stroma; glandular HOXA10 expression was unchanged.

Endometria from uteri with submucosal myomas also demonstrated a downward trend in BTEB1 mRNA expression, but not in LIF levels.

Clinical implications

Although submucosal myomas have been linked to poor reproductive outcomes, the histological evaluation of endometria from uteri with submucosal myomas did not reveal any specific defect that might impair embryo implantation. But subsequent molecular analysis suggests a mechanism by which submucosal myomas may disrupt reproduction. Our findings are consistent (CONTINUED ON PAGE 8)

New insights on the link between myomas and infertility

By Beth W. Rackow, MD

Dr. Rackow is Assistant Professor of Obstetrics, Gynecology and Reproductive Sciences and of Pediatrics at the Yale School of Medicine.

Yale Advancing Ob/Gyn          Spring 2011

4
Americans are victims of their own prosperity. We have such an abundance of inexpensive food that many of us find it next to impossible to stop eating once our daily caloric needs are met. The resulting obesity epidemic has contributed to a long list of chronic diseases and has cost us billions of dollars in health care expenditures.

No doubt the etiology of this epidemic is complex and includes cultural, physiologic and psychologic factors, many of which are beyond the control of individual clinicians. But there are several practical measures Ob/Gyns can take to manage reproductive-age patients who are already obese, and counsel those who are headed in that direction.

Yale researchers link obesity to cesarean delivery

Obesity is defined as a body mass index (BMI) above 30 kg/m² and overweight as 25 to 29.9 kg/m². In recent years, we at Yale-New Haven Hospital have delivered several patients with BMIs in excess of 80 kg/m²! The list of complications faced by pregnant obese patients is quite long and includes – but is certainly not limited to – increased risks of:

- Spontaneous abortion and neural tube defects
- Gestational hypertension, preeclampsia and gestational diabetes
- Fetal macrosomia and shoulder dystocia
- Postpartum venous thromboembolism

In recent years, Yale investigators have also been looking at the link between cesarean delivery (CD) and obesity. Urania Magriples, MD, and her colleagues studied over 800 pregnant women receiving group prenatal care in New Haven, Connecticut, and Atlanta, Georgia, and found that BMI was able to predict CD. After taking into account possible confounding factors, their multivariate analysis revealed that the odds of an obese woman delivering by cesarean section were more than twice those seen in normal-weight women (odds ratio 2.3, 95% CI 1.48 to 3.58, p<0.0001). Several possible mechanisms may explain the association: Obese patients have more soft tissue in the pelvis, which in turn narrows the pelvic outlet; their fetuses are larger; they may have a primary impairment in myometrial function since they require relatively higher doses of oxytocin to induce labor or maintain contractile force; and physicians may be more inclined to do a CD in these patients because of greater concern about macrosomia and shoulder dystocia.

Regardless of the mechanism of action, these patients need special attention. One of the first priorities in caring for a morbidly obese gravida is a “gap analysis” to determine if your obstetrical team has all the necessary equipment on hand, including oversize beds and wheelchairs. For all obese patients you should also consider enhanced antenatal surveillance for complications like diabetes and hypertension; early diabetes screening is one option, as are more frequent prenatal visits to detect the presence of preeclampsia. You’ll also want to have clearly delineated guidelines for intrapartum care: Palpating for contractions can be problematic in this patient population because of all the extra abdominal adipose tissue, and external fetal monitoring has its limitations in obese patients because of the distance between the skin and the uterus. Internal monitoring with intraventricular pressure catheters and/or fetal scalp electrodes may be justified if there are no contraindications to these modalities.

Don’t overlook the simple things

In our age of high-tech medicine, it is easy to overlook less sophisticated approaches to obesity. Such low-tech approaches can be especially helpful if you are fortunate enough to be caring for a patient before she conceives. Two of the mistakes some physicians make when caring for non-pregnant overweight patients are, first, the assumption that patients won’t pay attention to their

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Insights into embryo metabolism may help detect viable embryos (CONTINUED FROM PAGE 1)

**Understanding metabolomics**

A growing understanding of how embryos utilize nutrients and excrete metabolic byproducts before implantation is helping investigators determine which embryos are most viable, which in turn should reduce the number of embryos that need to be transferred to result in a live birth.

Over the years, we have learned that embryos in preimplantation culture have specific nutrient needs and that these needs change over time. Embryos survive *in vitro* by means of both aerobic glycolysis (the Krebs cycle) and anaerobic glycolysis (the Embden-Meyerhof pathway). Initially, as the embryo begins to develop, aerobic metabolism and pyruvate use predominate, but this pathway is gradually supplanted by anaerobic glycolysis using glucose, which predominates at the blastocyst stage. As the embryo moves through these phases of development, it also excretes specific metabolites related to the predominant metabolic pathway.

As the embryo’s nutrient consumption changes, these changes are reflected in the composition of the culture “soup.” The science of metabolomics measures these metabolic byproducts to help determine how viable the embryo will be once implanted.

**Clinical application**

Our research team at Yale has recently reported results from a multi-center study using a non-invasive spectroscopic method for prediction of the implantation potential of embryos in IVF. In this study, we showed that the metabolism of embryos that result in pregnancy is different than that of embryos that do not, and that the difference may be detected by the rapid, non-invasive evaluation of the embryo culture media using spectroscopic analysis and bioinformatics, also called metabolic profiling. The team evaluated spent culture media from transferred embryos using near-infrared (NIR) spectroscopy.

Our approach successfully predicted the implantation potential of embryos and was independent of morphology as a parameter. However, since traditional morphological embryo grading systems have merit, we wanted to determine if the two approaches would be of value when combined. With that in mind, we evaluated metabolic profiling as an adjunct to morphological grading systems. We found that when embryos were selected for implantation based on morphology alone, those with a higher metabolic-determined viability score were more likely to yield a successful pregnancy when compared to embryos with a low viability score.

Overall, our findings suggest that the addition of this technology to morphologic assessment may improve our ability to determine the viability of individual embryos. The use of such a rapid, non-invasive, objective and affordable technology has the potential to decrease multiple gestations and improve pregnancy rates in the near future.

**References**


Stem cell therapy suggests a profound role for endogenous stem cells

Research on the potential role of human stem cells as therapeutic agents is giving us new insights into the role that endogenous stem cells play in tissue repair and regeneration. The evidence strongly suggests that stem or progenitor cells exist in most tissues and may help repair damaged cells in a variety of chronic diseases, including neurodegenerative disorders.

Conventional wisdom says that neurons in the brain lose their ability to regenerate once the brain is fully developed, but we now know that neurons are actively involved in adult tissue remodeling. Such tissue activity suggests that stem cell therapy may actually be mimicking the body’s innate regenerative powers. In fact, the evidence even suggests that chronic diseases may occur as endogenous stem cells are depleted over a lifetime of tissue damage and repair. This theory would also explain why infants have such neural plasticity—their brains still have an ample supply of stem cells to regenerate damaged tissues—while older adults burdened by PD do not. In effect, their endogenous supply of stem cells has been “used up” over a lifetime of tissue repair.

References


New insights on the link between myomas and infertility (CONTINUED FROM PAGE 4)

with the accepted role of HOXA10 and HOXA11 in endometrial receptivity; the data also suggest that BTEB1, a downstream HOXA target gene, is involved in endometrial receptivity.

Our investigation has several implications. This is the first study to identify an endometrial abnormality that explains why poor reproductive outcomes are seen in women with submucosal myomas. Furthermore, the fact that the molecular abnormalities were present throughout the uterus and not just over the submucosal myoma means these tumors may affect the entire endometrial lining. Hence, the mechanism by which submucosal myomas impact endometrial receptivity is not simply a local mechanical effect over the myoma, but involves a signaling mechanism to the entire endometrium. Additionally, this widespread endometrial effect did not simply correlate with the size of the submucosal myoma. Therefore, submucosal myomas should be surgically resected prior to infertility treatment. Future studies may identify the signaling molecule involved in this pathway, which could serve as a novel therapeutic target for the treatment of infertility in women with myomas.

References


Practical advice on managing the obese gravis (CONTINUED FROM PAGE 5)

admonition to lose weight and exercise, and second, that there are no effective programs worth recommending.

Research data strongly suggest that patients do pay attention to physicians’ advice to change their lifestyle, as evidenced by the positive effect that telling patients to quit smoking has on smoking cessation rates. And while most Ob/Gyns don’t have the time or resources to provide in-depth weight loss counseling, they can refer the patient to nutritionists and psychologists who do have the time and expertise. The American Dietetic Association’s website has a nutritionist locator and can direct you to Registered Dieticians who specialize in obesity (www.eatright.org). Similarly, the American Psychological Association website’s practitioner locator will allow patients to locate clinicians who specialize in eating disorders (apa.org/).

References


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