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Ectopic Pregnancy

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An ectopic pregnancy occurs when an embryo implants outside of the uterus. In 98% of cases, the ectopic implantation takes place in the fallopian tube. Ectopic pregnancies occur in 1% to 2% of all pregnancies and remain an important cause of maternal morbidity and mortality in the first trimester. Although the etiology of ectopic pregnancy is poorly understood, epidemiologic studies have identified several risk factors for ectopic pregnancy: cigarette smoking, tubal damage from previous surgery, and *Chlamydia trachomatis* infection. These risk factors have been hypothesized to lead to embryo implantation within the fallopian tube by altering tubal smooth muscle contractility and the tubal microenvironment, leading to arrest of the embryo within the fallopian tube and an environment more apt to facilitate implantation.

Ectopic pregnancy can be difficult to diagnose, and most women present with pain and bleeding in the first trimester. These symptoms, however, are relatively common in early pregnancy, are not specific to ectopic pregnancy, and may be associated with other conditions, such as miscarriage. There are currently no specific biomarkers for ectopic pregnancy, and diagnosis relies on serial measurements of serum β-human chorionic gonadotropin (β-hCG)[9] to monitor the β-hCG doubling time, as well as transvaginal ultrasound (TVS), to rule out the presence of an intrauterine pregnancy. In some cases, serum β-hCG concentration and ultrasound results may be inconclusive, and laparoscopy is required to make a diagnosis. In this Q&A, 4 experts discuss recent advances that help us understand the etiology of ectopic pregnancy and the available methods for diagnosing and treating ectopic pregnancy.

How is ectopic pregnancy currently being diagnosed in your institution?

Andrew W. Horne: In our institution, ectopic pregnancy is diagnosed with a combination of TVS and serial serum β-hCG monitoring.

Kurt Barnhart: The majority of women with ectopic pregnancy are identified with ultrasound. Some women will be identified as having an ectopic pregnancy, and others will be identified as having an intrauterine pregnancy, therefore virtually eliminating the possibility of concomitant ectopic pregnancy. However, there remain up to 20% of women at risk who have nonspecific ultrasound findings, and further testing is warranted to ultimately distinguish the location of the gestation. This follow-up usually occurs as an outpatient.

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[9] Nonstandard abbreviations: β-hCG, β-human chorionic gonadotropin; TVS, transvaginal ultrasound; PUL, pregnancy of unknown location.

Tom Bourne: In our unit, the majority of ectopic pregnancies are diagnosed with TVS. We can detect 74% of ectopic pregnancies on the basis of a single TVS. After follow-up scans, 91% of ectopic pregnancies can be visualized before surgery. The reasons some ectopic pregnancies are not seen initially are that the fetuses are simply too small and it is too early in the disease course for them to be visualized. A proportion of women undergo a TVS, and still the location of the pregnancy cannot be identified. This pregnancy is classified as a pregnancy of unknown location (PUL). In these circumstances, serial serum β-hCG concentrations are measured to determine the intensity of follow-up, and further scans are arranged until the location of the pregnancy is identified. In the event that the serum β-hCG continues to rise and the rate of rise is too slow to be associated with a viable intrauterine pregnancy, a presumptive diagnosis of ectopic pregnancy may be made, and medical treatment with methotrexate is begun. Laparoscopy is rarely used to make a diagnosis of ectopic pregnancy but is used for treatment. If a laparoscopy is carried out in our institution on the basis of the prior visualization of an ectopic pregnancy with TVS and no ectopic pregnancy is then seen at surgery, this would be considered a risk issue and a potential indicator of there being a problem with the quality of the scan that had been carried out.

Ioannis E. Messinis: The diagnosis of ectopic pregnancy is straightforward only in the case of rupture, with symptoms of acute abdominal pain and hemodynamic shock. In that case, history and physical examination are enough for the diagnosis. In the case of a suspected ectopic pregnancy without symptoms, the diagnosis is based on history, physical examination, TVS, and serum β-hCG measurement. The limitations of all these methods are well known. An ectopic pregnancy is highly suspected if the serum β-hCG is >2000 IU/L with no sac in the uterine cavity on ultrasound and is diagnosed by laparoscopy. When the serum β-hCG on the first visit is <2000 IU/L, the doubling time of β-hCG values is estimated. In these cases, methotrexate, uterine curettage, and laparoscopy are considered for treatment.

What are the difficulties in diagnosing ectopic pregnancy? How do these affect patient care?

Andrew W. Horne: The diagnosis of ectopic pregnancy remains problematic and often results in treatment delays. In our unit, fewer than 50% of tubal ectopic pregnancies are diagnosed at the patient's initial presentation. Despite the clinical advances in imaging, ultrasound is nonconclusive in up to 20% of women, for whom measurement of serial β-hCG concentrations is necessary to guide management. Further difficulties are encountered because serial β-hCG determination cannot accurately separate arrested intrauterine from ectopic pregnancies. Decelerated increases in β-hCG concentrations cannot be used to discriminate between a miscarriage and an ectopic pregnancy. Moreover, laparoscopy can be occasionally required to confirm the diagnosis, and this procedure is not without risk to the patient.

Kurt Barnhart: Ectopic pregnancy, if not identified or considered, can be a life-threatening event. However, clinicians are now very good at identifying women at risk, and the clinical dilemma has shifted from the possibility of missing an ectopic pregnancy to determining the acuity of follow-up and/or interventions. Paramount is the determination that a woman at risk for an ectopic pregnancy does not have a desired viable intrauterine pregnancy. This is necessary because the diagnosis and treatment of ectopic pregnancy will often result in termination of a pregnancy. The second step is to distinguish if a nonviable pregnancy is an ectopic pregnancy or a miscarriage.

Tom Bourne: In developed countries, most ectopic pregnancies will be diagnosed before there are serious acute complications. However, the problem is with diagnosing ectopic pregnancy at a point in the disease process when relatively conservative treatment approaches remain an option. As indicated earlier, at a certain point ectopic pregnancies are simply too small to visualize with current imaging techniques. Furthermore, at least 10% of early ectopic pregnancies have a rise in β-hCG similar to that seen in a normal viable intrauterine pregnancy. Using the β-hCG ratio is certainly not diagnostic and only can help us focus on a PUL that requires a closer follow-up until the location of the pregnancy is known. We can use progesterone, but while this might help with viability, it does not inform us about pregnancy location. So the problem is in diagnosing ectopic pregnancies at a very early stage when treatment is likely to be straightforward and not

involve surgery or require treatment at all. The use of laparoscopy as a diagnostic test is a very invasive approach and should be unnecessary in most cases, and even when a laparoscopy is performed, it may be impossible to visualize a small ectopic pregnancy within a fallopian tube.

More recently, the problem of diagnosing "nontubal" ectopic pregnancy has become more of an issue. By this I mean pregnancies that have implanted outside the endometrial cavity but not in the tube. Of particular concern are pregnancies that are implanted in cesarean section scars. In the early stages, these pregnancies are relatively easy to treat, but if advanced, they are associated with significant morbidity and mortality. The ultrasound-based diagnostic criteria for these pregnancies are not clear yet, and the risk of a false-positive diagnosis and intervention is not known.

Ioannis E. Messinis: Apart from the usual clinical manifestations of the normal pregnancy, there are no specific symptoms associated with ectopic pregnancy. In some cases, mild abdominal pain and vaginal bleeding are present. Except in the case of acute abdominal pain and hemodynamic shock, a physical examination is not diagnostic, and the ectopic sac is rarely visible on ultrasound. An ectopic pregnancy is likely if there is less than a 66% rise in β-hCG values over 48 h. Nevertheless, even with stable values, at a β-hCG concentration $<$2000 IU/L the differential diagnosis from a missed or incomplete abortion is difficult. In the majority of cases, the ectopic pregnancy is a diagnosis of exclusion. As a result, a patient's treatment may be delayed, which may predispose to the rupture of the ectopic, while in other cases a precipitate action may lead to an unnecessary surgical intervention. This might cause substantial psychological morbidity.

What would an ideal biomarker for ectopic pregnancy look like?

Andrew W. Horne: An ideal serum biomarker would be one that could be assayed accurately and quickly, preferably in an emergency department setting. However, it would also have to be an inexpensive measure to have true value clinically.

Fundamentally, the question of whether a serum biomarker exists that can accurately and specifically detect a tubal ectopic pregnancy remains unanswered. Furthermore, with the advent of better imaging techniques, a serum biomarker may be superseded by ultrasound-related technology.

Kurt Barnhart: An ideal biomarker needs to be noninvasive and highly accurate. However, the test characteristics of the biomarker can certainly be taken advantage of, as there could be a variety of uses. For example, a valuable biomarker could distinguish between an intrauterine and an extrauterine (ectopic) pregnancy. Alternatively, a biomarker could also be valuable if it could distinguish between a viable and a nonviable pregnancy, especially when the location of that pregnancy is not known. Finally, a biomarker could also be used to identify the aggressiveness and prognosis of a PUL, distinguishing a patient who may be treated with expectant management from a patient best suited for either medical or surgical therapy.

Tom Bourne: An ideal "fantasy" biomarker would have a number of positive characteristics. First, the specimen required for testing would be simple to collect. Ideally, it would be a urine-based test that could be carried out by the pregnant women at home as a screening test. If a serum-based test, it would be helpful if the test could be carried out in the clinic where the patient is seen so the patient could be given immediate instructions without the delay involved in sending samples to a laboratory. A focus on diagnostic sensitivity would be needed for such a biomarker. Another important function of such a marker would be to predict the behavior of the ectopic pregnancy. Will the ectopic pregnancy fail? Will it respond to methotrexate or a future substitute? Will the pregnancy end with tubal rupture and hemorrhage? A marker that could help predict these outcomes (and monitor treatment) would significantly reduce the morbidity of ectopic pregnancy. It is likely that many ectopic pregnancies are currently treated medically or surgically when they would resolve without any intervention. The problem with using β-hCG concentration change over time as a predictor of ectopic behavior is that we are observing the pregnancies only over a limited period of time in their natural history. The β-hCG concentration may rise for a few days and then decline. However, with current approaches, intervention will probably occur before any decline in β-hCG concentration is observed.

Ioannis E. Messinis: An ideal biomarker would be a protein secreted exclusively by the ectopic trophoblast and easily measurable in blood. Until now, apart from β-hCG, other substances that have been evaluated for the diagnosis of an ectopic pregnancy include pregnancy-associated plasma protein A, vascular endothelial growth factor and its soluble receptor, placental growth factor, activin A, human placental lactogen, and serum-specific protein 1. Also, factors produced by the corpus luteum, (e.g., progesterone and inhibin A) or endometrial proteins (e.g., leukemia inhibitory factor and glycodelin) have been included. Nevertheless, none of these substances is considered an ideal biomarker for ectopic pregnancy.

Is the difficulty in diagnosing ectopic pregnancy adding substantial financial burden to the healthcare system, and would a new biomarker improve this?

Andrew W. Horne: The inevitable multiple visits and tests currently necessary are a sizeable expense for health services. As an example, we refer to data from a recent study performed in Edinburgh that indicate that health services in Scotland are spending up to £1.5 million (US$2.37 million) per year diagnosing and excluding ectopic pregnancy, with an estimated £9 million (US$14.22 million) in direct costs alone to health services per year throughout the United Kingdom. Using a theoretical diagnostic serum biomarker, we calculated that we could save health services up to £1 million (US$1.58 million) every year in Scotland.

Kurt Barnhart: Currently, when the location of a gestation is not identified by ultrasound, a woman needs repetitive interactions with the healthcare system. These include diagnostic tests, additional ultrasounds, and, at times, presumptive treatment. This adds tremendously to the burden on the healthcare system, especially when a woman presents to a different clinical entity and many of these tests are repeated. A biomarker that would accurately identify the location of a pregnancy, the viability of the pregnancy, or the prognosis of the pregnancy would immediately shorten the number of visits and time to diagnosis, thus greatly reducing costs.

Tom Bourne: Undoubtedly, there is a large financial burden associated with this problem. The issue relates principally to a PUL; between 10% and 30% of women presenting with problems in early pregnancy will be classified as such. Many women undergo multiple blood tests and/or ultrasound scans before a definitive diagnosis is made. Other than a healthcare cost, there is also a cost to the women themselves and society in terms of time off work and child care. The psychological morbidity of such protracted follow-up while being uncertain about the outcome is not known but may be great. Perversely, the introduction of sensitive home pregnancy-testing kits has also exacerbated the problem. Women now may know they are pregnant before they have even missed a period. As a result, we are seeing people come for scans earlier in gestation, either for reassurance or with symptoms when there is no possibility that a pregnancy will be seen by ultrasound. Inevitably, they are then classified as a PUL and may end up having a number of unnecessary tests as a result. The relationship between the likelihood of an inconclusive scan and the gestation when the scan is carried out was nicely shown in a paper by Cecilia Bottomley.

A further cost relates to a late or missed diagnosis of ectopic pregnancy. This cost may be in terms of avoidable surgery and hospitalization, or the impact on future fertility. Unnecessary treatment with methotrexate is also a real issue. The inappropriate administration of methotrexate to women with a PUL who are found subsequently to have a viable intrauterine pregnancy is an increasing cause of litigation, with its associated costs.

Ioannis E. Messinis: The methods used for the diagnosis of an ectopic pregnancy are time-consuming. It is estimated that no more than 50% of the cases of ectopic pregnancies are diagnosed at the first visit. On the other hand, the biomarkers used so far have a low diagnostic sensitivity and diagnostic accuracy, leading to a long process of investigation with the need for either multiple hospital visits or even an extended hospital stay. Furthermore, on several occasions an unnecessary laparoscopy can be performed, which carries an increased risk for the patient. All of these situations add a substantial financial burden to the healthcare services that are extremely important, particularly for countries with limited resources. The discovery of a new biomarker specific for the ectopic pregnancy would certainly reduce these costs.

In your opinion, what factors have hindered the discovery or validation of an ectopic pregnancy biomarker?

Andrew W. Horne: Over 20 serum biomarkers have been identified to date in an attempt to permit earlier diagnosis of ectopic pregnancy, the instigation of earlier management, and a reduction in healthcare costs. The clinical utility of these biomarkers has been limited because of variable results due, for the most part, to limitations in study design. In many studies, the cohort examined was very small, and the prevalence of ectopic pregnancy within the study population was not constant. In some studies, patients were not accurately matched for gestation. This limitation reflects the difficulty in determining the gestational age of an ectopic pregnancy. Some of the serum biomarkers have also limited their own use because they did not follow a steady pattern (increase or decrease) with a normal gestation. Moreover, changes in the assays and the reagents used to detect biomarkers have led to conflicting results between studies.

Kurt Barnhart: There have been a number of studies identifying putative biomarkers for ectopic pregnancy. The main problems have been small study sample sizes and lack of follow-through and validation. Often, authors are content with a peer-reviewed publication. Since there is a bias toward positive findings in publication, often these findings are overly optimistic and not reproducible. What is needed is a concerted effort

to pare down purported biomarkers and to initiate large-scale studies to validate these biomarkers, preferably in a population separate from the ordinal development. The final step will be to determine their optimal use in clinical practice.

Tom Bourne: One of the issues is the age-old need for close collaboration between busy "high turnover" clinical units and laboratory research. A minimum investment in structure to ensure well-categorized samples that are collected from the right patients is money well spent. A lack of such infrastructure has meant that many studies are based on small numbers of subjects with relatively few ectopic pregnancies among them. To give this some perspective, in our unit we need to see about 10 000 women in early pregnancy to generate 1000 PULs. From 1000 PULs, we will have at most 100 ectopic pregnancies. Close collaborations between clinicians and researchers are needed to ensure that all available tissue samples end up in the research laboratory.

Ectopic pregnancies are also a heterogeneous group: different or unknown gestation, baseline β-hCG, β-hCG ratio, serum progesterone, maternal age, failing, viable, etc. The result is that research in this area has not always been based on sufficient numbers of well-characterized samples. We need to make sure there are research networks that ensure good links between clinical services, where samples are collected and outcomes are recorded, and the laboratory.

An important issue is also nomenclature. For example, the definitions of a PUL differ between countries and even between institutions. If we cannot agree on nomenclature, we will always find it hard to conduct meaningful research.

We have also been hampered by a failure of clinicians to appreciate that "overtreatment" of ectopic pregnancy is unacceptable; it is not a good idea to carry out unnecessary procedures or treatments on people! The attitude now has shifted from telling the patient, "you are so lucky we saved your life," to recognizing that the woman has lost a pregnancy and that treatment, if necessary, should cause as little psychological and physical harm as possible. This has now led to clinicians and patients recognizing that there would be benefit from better biomarkers, both for earlier diagnosis and for monitoring treatment.

Ioannis E. Messinis: In my view, this is related to the lack of specific substances secreted from the ectopic trophoblast that would be unique to this tissue and not produced from the eutopic trophoblast. Certainly, future research may provide valuable information on this matter. An interesting hypothesis would also be the production of a specific substance from the tubal mucosa following activation by the implanted blastocyst. Although the majority of ectopic pregnancies occur in the fallopian tube, a marker produced from the tubal mucosa will not cover all other pelvic and abdominal locations, such as the ovaries or the abdominal cavity, where ectopic implantation may also occur. Furthermore, large prospective randomized trials are needed to evaluate the effectiveness of a combination of factors in the context of a predictive model for an early diagnosis of the ectopic pregnancy.

Very little is known about the etiology of ectopic pregnancy. What obstacles are inhibiting this research?

Andrew W. Horne: Prior studies on the etiology of ectopic pregnancy have been largely descriptive and focused on dysregulated gene or protein expression, comparing fallopian tubes collected from women with ectopic pregnancy and fallopian tubes collected from nonpregnant women or women with a "pseudopregnancy." There has been very little analysis of the functional consequences of the observed changes in gene or protein expression reported in these studies, largely due to the fact that the etiology of ectopic pregnancy is difficult to study. There are no suitable animal models, because ectopic gestation is rare in animals. Ethical constraints inhibit the collection of fallopian tube biopsies from women with healthy intrauterine pregnancies, making it difficult to obtain the ideal control for comparison to fallopian tubes from women with ectopic pregnancy. In addition, researchers must rely on information derived from fallopian tube biopsies obtained from women with ectopic pregnancy rather than before the event. It is difficult to ascertain whether the molecular changes observed predispose to ectopic pregnancy, or whether they are simply the result of tubal implantation and/or the presence of the embryo. Furthermore, although the epidemiological risk factors for ectopic pregnancy (e.g., chlamydial infection, cigarette smoking) have been well documented, the exact mechanism by which infection or smoking leads to tubal implantation remains unexplained.

Kurt Barnhart: The study of human pregnancy is very difficult for ethical reasons. While there is an epidemiologic study assessing risk factors for ectopic pregnancy, it is difficult to identify causal factors. Moreover, we have large gaps in our knowledge regarding our understanding of normal implantation, as well as ectopic implantation. We also do not understand how the risk of ectopic pregnancy is modified by concomitant disease, age, or subfertility. Finally, the lack of animal models has hindered the ability to research this disease.

Tom Bourne: Some of the risk factors, such as smoking and chlamydial infection, are known, but the mecha-

nisms whereby these factors lead to tubal pregnancy remain unclear. The presumption is that studies of the fallopian tube will shed light on this. However, studies are often limited to examining the fallopian tube of women where the tube has been removed because of an ectopic pregnancy, which means any findings are difficult to interpret. In these circumstances, biopsy of the contralateral tube is not ethical. Again, there are often difficulties in organizing sample collection. Cases of interest would perhaps be hydrosalpinges removed following pelvic infection or before in vitro fertilization, or the fallopian tubes of women who smoke or who have had *Chlamydia* and are undergoing laparoscopic sterilization.

The mechanism behind nontubal ectopic pregnancy is not understood but may relate to the healing of cesarean section scars and, in the case of cervical and myometrial pregnancy, to instrumentation of the cavity. A lack of agreed-upon nomenclature to describe section scars has been a problem when trying to ascertain whether the ultrasound appearances of a scar relate to healing and/or the risk of ectopic pregnancy.

Ioannis E. Messinis: Various clinical conditions have been associated with the occurrence of an ectopic pregnancy, such as previous pelvic inflammatory disease, previous tubal pregnancy, previous tubal surgery for infertility, and current use of an intrauterine contraceptive device. In the majority of the cases, damage of the tubal mucosa can explain the ectopic implantation. Additional risk factors include infertility treatment, such as ovulation induction and in vitro fertilization. An explanation of the implantation at locations other than the fallopian tube is difficult. Research, however, is limited, and this is possibly due to the rarity of these locations, since each of them (cervix, abdomen, ovary) accounts for <1% of ectopic pregnancies. On the other hand, tubal infection, the most common cause of mucosal damage predisposing to an ectopic pregnancy, may recur even after treatment. Permanent scarring of the tube is not unusual.

How are ectopic pregnancies managed at your institution, and would an early-detection marker change the management of this condition?

Andrew W. Horne: In Edinburgh, ectopic pregnancy is managed expectantly, medically (with methotrexate), or surgically. Women managed expectantly have careful monitoring and assessment to establish whether the ectopic pregnancy will resolve without the need for intervention. This conservative approach is used only if the patient is stable, is asymptomatic, has a low risk of rupture, and has decreasing serum β-hCG concentrations. Medical management of ectopic pregnancy is offered to patients with minimal symptoms who are hemodynamically stable, have no more than a moderate amount of intra-abdominal free fluid on ultrasound scan, and have a β-hCG concentration <3000 IU/L. Medical management involves the intramuscular administration of methotrexate, usually in a single dose. After receiving methotrexate, patients undergo close monitoring until their serum β-hCG concentration drops below 5 IU/L. Surgical management is indicated if the patient is not eligible for methotrexate or has symptoms or signs of tubal rupture. This involves a salpingectomy if the contralateral tube is healthy or a salpingotomy when this is not the case. In the absence of acute hemodynamic compromise, a laparoscopic (keyhole) approach is used.

Unfortunately, the efficacy of medical management with methotrexate falls with increasing ectopic size, and this means only 25% of women with ectopic pregnancies are suitable for medical management in Edinburgh. An early-detection marker would allow a greater number of women to have successful medical management.

Kurt Barnhart: Currently, most ectopic pregnancies are managed with the chemotherapy agent methotrexate to medically treat the ectopic pregnancy. In fact, this treatment is often administered to women with a suspicion of, but without confirmation of, a diagnosis of ectopic pregnancy. Surgical management has become less common. However, surgical management of an ectopic pregnancy has some strong benefits, including the swiftness and definitiveness of its treatment. Surgery also allows a better assessment of prognosis for the next pregnancy. Women treated with medical therapy need continued outpatient surveillance until the gestation has resolved. This can take up to 6 weeks. If a biomarker could accurately identify the location of a gestation, we could have individualized and targeted therapy. Additionally, if a biomarker could assess prognosis, a greater number of woman might be able to be treated expectantly, therefore reducing the morbidity from both the medical and surgical therapies involved.

Tom Bourne: A mixture of surgery, medical treatment with methotrexate, and expectant management (watch and wait with no treatment). Surgically, approximately 95% of tubal ectopic pregnancies are operated on laparoscopically (keyhole surgery), and a salpingectomy or salpingotomy is performed, depending on the amount of damage to the affected tube and the state of the contralateral tube. Laparoscopy can be carried out even in the event of hemodynamic compromise, but laparotomy may be necessary in these circumstances, depending on the skill of the surgeon. Patients are selected for surgery if they are clinically unstable, if they have a large

hemoperitoneum according to ultrasound, if an embryo with a heartbeat is visible within the gestation sac of the pregnancy, or if the ectopic mass is very large.

Conservative management strategies can be considered if the patient is compliant and there are robust procedures in place for follow-up. In general, we use the β-hCG ratio to select women for expectant management (generally a declining β-hCG) with methotrexate. Following treatment with methotrexate, the patients are followed up with the serum β-hCG concentration taken on days 4 and 7 according to the original protocol by Stovall. We use a single-dose protocol.

Biomarkers for ectopic pregnancy could greatly impact management. Earlier diagnosis could make more women candidates for medical management, which might involve lower doses of methotrexate or other combinations of drug therapy. There is also a view that many women receive methotrexate unnecessarily for ectopic pregnancies that would resolve without intervention if allowed to evolve naturally. Currently, an increasing β-hCG concentration in general will lead to methotrexate treatment rather than expectant management. Clearly, in some of these pregnancies, the β-hCG will subsequently fall, the pregnancy will fail without the need for treatment, and we are simply observing the pregnancy at a single point in its natural history. We see this after methotrexate, where in the 4 days after treatment there is often an alarming rise in serum β-hCG, only for it to fall again between days 4 and 7. So, a biomarker that would predict the failure of an ectopic pregnancy, the risk of tubal rupture, and the response to methotrexate or other drug therapy would lead to changes in management for a number of women. The use of such a marker would improve patient selection and allow us to better individualize treatment.

Ioannis E. Messinis: We measure β-hCG and calculate the doubling time. We also use ultrasonography to exclude or diagnose an intrauterine pregnancy. If the β-hCG concentration is >2000 IU/L and no intrauterine sac is visible, we do a laparoscopy. If the β-hCG concentration is <2000 IU/L with at least a 66% rise over 48 h, the patient is followed up until the β-hCG concentration rises above that level. If there is little or no increase and no intrauterine sac is seen, methotrexate treatment or laparoscopy plus uterine curettage are discussed with the patient, unless vaginal bleeding starts and β-hCG concentrations decrease. In these cases, uterine curettage may be needed. The availability of an early-detection marker would certainly help prevent the rupture. This is expected to reduce the number of visits and the financial burden. It is also expected that the use of medical treatment (methotrexate) will increase at the expense of surgery.

Are there any precautions a woman can take to avoid having an ectopic pregnancy?

Andrew W. Horne: Although women with ectopic pregnancy frequently have no identifiable risk factors, it has been shown that increased awareness of ectopic pregnancy and a knowledge of the associated risk factors help identify women at a higher risk to facilitate an early and more accurate diagnosis.

Most risk factors are associated with risks of prior damage to the fallopian tube. These factors include any previous pelvic or abdominal surgery, and pelvic infection. *Chlamydia trachomatis* has been linked to 30%–50% of all ectopic pregnancies. The exact mechanism of this association is not known, but it has been proposed that in addition to distortion of tubal architecture, it may be due to an effect on the tubal microenvironment.

In addition, one-third of all cases of ectopic pregnancy are thought to be associated with smoking. There is a dose–effect relationship, with the highest adjusted odds ratio of 3.9 when >20 cigarettes are smoked a day. Several mechanisms for this association have been suggested, including one or more of the following: delayed ovulation, altered tubal and uterine motility and microenvironment, and altered immunity. Thus, women should be advised to seek early treatment for suspected pelvic infections and to stop smoking.

Kurt Barnhart: Unfortunately, there are no known preventative measures for ectopic pregnancy. At least half of all ectopic pregnancies occur in women without any risk factors. To avoid the complications of ectopic pregnancy, it is best for a woman to recognize the signs and symptoms of a potential ectopic pregnancy, which include abnormal vaginal bleeding and pelvic pain in the first trimester of pregnancy.

Presentation to an astute clinician will allow early diagnosis of a potential ectopic pregnancy. However, it should be noted that many women present so early in a gestation that diagnostic tests have lower accuracy, and an intervention is often suggested by the healthcare system prematurely. A clinician who understands the pathophysiology of ectopic pregnancy can appropriately follow a woman, and intervention and misdiagnosis can be minimized.

Tom Bourne: Clearly, avoiding the risk factors would be a move in the right direction. Not smoking. Using appropriate contraception may seem obvious but needs to be emphasized to some of our patients. A greater emphasis on sexual health is important, especially in young women. Using barrier methods of contraception (maybe in addition to the pill) will reduce the risk of *Chlamydia* and other sexually transmitted

infections. Screening for *Chlamydia* may lead to earlier treatment, although whether this reduces the risk of ectopic pregnancy or other reproductive complications is not certain. Clinicians and women need to have a high index of suspicion for *Chlamydia* infection, and clinicians need to consider the diagnosis for any young woman with abnormal bleeding or pain. The fact that a recent paper stated that 17% of 70 000 Nordic women 18–45 years old reported having had a *Chlamydia* infection suggests the situation could be improved.

Women at risk of ectopic pregnancy should also be made aware of the risk and be encouraged to seek an early scan to identify the location of the pregnancy. If an ectopic pregnancy cannot be avoided, it may at least be diagnosed early.

It is hard to give advice regarding reducing the risk of nontubal ectopic pregnancy. Clearly, having had a cesarean section is a risk factor and should be considered as a potential future risk for any women electing to deliver this way.

Ioannis E. Messinis: No specific precautions exist to avoid ectopic pregnancy. What really matters is to prevent serious complications by an early diagnosis and treatment. Therefore, women with one or more risk factors for an ectopic pregnancy should be closely monitored during the first weeks until an intrauterine sac becomes visible on ultrasound. Nevertheless, safe sex measures, including using a condom every time, help prevent sexually transmitted diseases such as *Chlamydia*. Smoking predisposes women to ectopic pregnancy. Therefore, women should be advised to quit smoking before and during pregnancy.

In this era of genomic medicine, have any genomewide association studies been performed to identify potential genetic factors linked to ectopic pregnancy?

Andrew W. Horne: To my knowledge, no genomewide association studies linked to ectopic pregnancy have been performed. However, we have shown in a small population that women with ectopic pregnancy exhibit differences in the distribution of single-nucleotide polymorphism alleles of the *CNR1*[10] [cannabinoid receptor 1 (brain)] gene, encoding the endocannabinoid receptor CB1. Replication of these data in a larger sample is needed to reach the conclusion that the *CNR1* gene, and indeed CB1, has a role in ectopic implantation.

Kurt Barnhart: My understanding is that there have been no genomewide association studies performed to identify genetic factors linked to ectopic pregnancy. However, it should be noted that genomewide association studies have not been as productive as hypothesized, and few have affected medical care to date. Perhaps rather than searching for a genetic predisposition, which may or may not be realized, we should be looking for a proteomic predisposition for the early diagnosis of ectopic pregnancy. In other words, it doesn't do us much good to find that somebody has a genetic predisposition when the condition is still a rare event (only 1%–2% of pregnancies), whereas a change in a woman's proteome that alerts a clinician to an abnormal pregnancy or an ectopic pregnancy would be of much greater value to an individual woman and to the healthcare system.

Ioannis E. Messinis: So far, there are no such studies in ectopic pregnancy. There are only some data on polymorphisms of specific genes. For example, although serum vascular endothelial growth factor concentrations are usually increased in ectopic pregnancies, no association between polymorphisms of the vascular endothelial growth factor gene and ectopic pregnancy was found recently. In addition, certain mannose-binding lectin genotypes have been associated with tubal damage in patients infected by *Chlamydia trachomatis*, a potential cause of ectopic pregnancy. Earlier data had suggested a possible association between oocyte chromosomal anomalies and ectopic pregnancy, but this has not been repeatedly examined. In addition, no association has been demonstrated between male factor infertility and ectopic pregnancy in in vitro fertilization.

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[10] Human genes: *CNR1*, cannabinoid receptor 1 (brain).