Epidemiology of Recurrent Pregnancy Loss

Spontaneous abortion (SAB) is defined as the expulsion of a fetus before 20 weeks of gestation or weighing less than 500 g.' Losses after 20 weeks are described as stillbirths or premature births and, generally, have different etiologies than losses earlier in gestation. The vast majority of SABs occur before 12 weeks of gestation. Those occurring at less than 8 weeks often do not contain an embryo and are sometimes referred to as "blighted ova. Recurrent abortion (RAB) is frequently defined as the occurrence of three or more consecutive SABs, although some of the case series of RAB have included women with two consecutive losses. The rationale for distinguishing RAB is based on the assumption that RAB may have an etiology distinct from that of a "sporadic" SAB.

In considering the topic of RAB from an epidemiologic standpoint, a critical issue is whether only studies of women with RAB should be included or whether studies of SAB are also relevant. A key premise of this article is that risk factors for RAB must be understood in the context of risk factors for SAB. Thus, we believe that both types of studies are relevant; and we will address the following questions. First, what types of epidemiologic studies are used to address SAB or RAB? Second, what is the probability of SAB in any given pregnancy and how is that risk affected by the outcome of the previous pregnancy? Third, what are the risk factors for SAB in general and for RAB in particular? Finally, on the basis of our review, how can epidemiologic studies of the causes and prevention of RAB be improved?

TYPES OF EPIDEMIOLOGIC STUDIES

The types of epidemiologic studies addressing SAB or RAB found in the literature include simple descriptive studies or case series, case-control studies, cohort studies, clinical trials, and meta analyses. We will briefly review the design of each of these types of studies and comment on their major strengths and weaknesses.

Case Series

A frequent type of study addressing SAB or RAB is the case series, describing characteristics of women with pregnancy losses.[2-6] The case series may provide statistics about the prevalence of a particular characteristic in selected populations. The usefulness of any particular statistic from a case series depends upon the sample size, which affects the precision of the estimate, and the demographics of the study population, which affects the generalizability of the findings.

The Case-Control Study

A case-control study is an extension of the case series in which a control group is added. Populations defined by the presence or absence of a condition, in this case SAB or RAB, are identified; and exposures prior to the SAB are then assessed, retrospectively, in both groups. The odds that cases were exposed to a particular agent divided by the odds that controls were exposed yields a measure called the exposure odds ratio, which describes the association between the exposure and SAB or RAB. In an unbiased study, an exposure odds ratio that is significantly greater than one (i.e., the null value) indicates that the exposure is associated with an increased risk for the condition. An important element of the study design is selection of a control group, with the logical choice for a study of SAB or RAB being those who successfully completed a pregnancy. In case-control studies, it is possible that cases may more readily recall or report a particular exposure than the control group, which would yield a falsely elevated relative risk associated with the exposure. Confounding may also occur when there is an imbalance in the distribution
of a variable between cases and controls that is associated with both the exposure and the condition. Age, as a known predictor of risk for SAB, would need to be carefully matched or otherwise adjusted for in a case-control study of SAB or RAB.

The Cohort Study

A cohort study starts with populations identified by the presence or absence of a particular exposure and follows the cohort members for the occurrence of an event, in this case, an SAB.~~[7] The rate at which the event occurred in the exposed population divided by the rate at which the event occurred in a nonexposed population is called the relative risk. As with the exposure odds ratio, a result significantly greater than one indicates an increased risk for the event associated with the exposure. Cohort studies can be prospective in nature (i.e., the exposure is assessed by the investigator, who must then wait for outcomes of interest to occur) or retrospective if the study is conducted from medical records of prior exposures and subsequent events that have already occurred. The ideal cohort study of SAB would begin even before the woman becomes pregnant so that careful and uniform monitoring could identify losses at the earliest gestations. In general, cohort studies are less likely to suffer from selection or recall bias but they are still subject to confounding, similar to case-control studies.

Clinical Trial

The clinical trial is a prospective cohort study that follows individuals who have been assigned a treatment to assess whether risk for a particular condition, in this case SAB, is reduced. The ideal clinical trial is one in which there is a placebo control, the treatment is randomly assigned, both subjects and observers are "blinded" as to treatment assignment, and the outcome is objectively assessed. These features minimize bias and provide strong evidence on the benefits (or risks) of a given treatment.

Meta-Analysis

The meta-analysis is not a distinct type of epidemiologic study but rather an analytic technique for combining the results from independent studies examining the same exposure (or treatment) and outcome so that a more precise estimate of the effect and powerful statistical test can be conducted.

A weighted average of the exposure odds ratios or relative risks from each of the studies is calculated with the weight inversely proportional to the variance of the association. This means that studies with larger sample sizes will contribute greater weight to the summary relative risk. As part of the analysis, a test for heterogeneity may be conducted; that is, whether individual study results are consistent or inconsistent with the summary relative risk. In relation to the topic of SAB and RAB, meta analyses of clinical trials examining various types of therapies have been conducted.[8]

RISK OF SPONTANEOUS ABORTION

The best source of information about the frequency of SAB comes from prospective studies of women who have discontinued a contraceptive method in an effort to conceive. Obviously, this is a select group of women with family planning goals and a willingness to be studied; yet only this type of study can yield information on both recognized and unrecognized pregnancy losses. In a landmark study, Wilcox et a1[9] collected daily urine specimens from 221 healthy women who were attempting to conceive and used a sensitive assay for human chorionic gonadotropin (hCG) to detect pregnancy. The assay results were not available to women or their physicians, who relied on the usual clinical means to diagnose a pregnancy. The probability of conceiving a clinically recognized pregnancy within a given cycle (fecundability) was about 250/o but was closer to 32o/o when unrecognized pregnancies were counted.

About 120/o of all clinically recognized pregnancies ended in an SAB. Among all pregnancies, including those detected only by assay, the total pregnancy loss was 3lo/o (including both recognized and unrecognized losses) and 22~/o for the unrecognized losses alone. The study also addressed the important clinical question of whether unrecognized losses contribute to clinical infertility; that is, are infertile women actually experiencing recurrent unrecognized losses? This did not appear to be the case: among the 14 women without a clinical pregnancy, the rate of unrecognized losses involved only 3o/~ of their cycles, whereas among the 40 women with unrecognized losses, 950/o

had a clinical pregnancy within 2 years.[9]

The most recent paper from the study'[0] reported that early losses were more likely the longer the interval between ovulation and the first elevation of hCG, which was correlated with implantation. Conceptions were most Likely to be successful when implanted 8—10 days after ovulation and less likely after 10 days. Theoretically, delays could re sult from late timing of intercourse leading to fertilization of an "older" ovum, delayed tubal transport, or delayed uterine receptivity. It is not clear, nor did the authors speculate, whether their observation might relate to "luteal phase defects" as a cause of RAB.

In another frequently cited longitudinal study, Regan et al" followed 630 women from the general population intending to become pregnant. As soon as a pregnancy was suspected or any symptoms of threatened abortion were seen, serial ultrasound studies were performed to document viability or loss. Among the 412 clinically recognized pregnancies, there were 357 successful pregnancies, 50 SABs, 2 ectopic (tubal) pregnancies, and 3 induced abortions. Excluding ectopic pregnancies and induced abortions, the overall occurrence of recognized SAB was 12%, matching precisely the frequency observed by Wilcox et al.[9] About half of the losses occurred prior to 8 weeks and only two of the losses occurred after 12 weeks of gestation.

Other important observations from Regan's study related to obstetrical history as a predictor of SAB. Women in their first pregnancy, women whose only other pregnancy was terminated, or women whose only or last pregnancy had been successful had a 4~6O/o chance of an SAB, with the 40/o pertaining to women whose pregnancies had all been successful. Alternatively, women whose only or last pregnancy had been lost had a l9~24O/o chance of an SAB, with the 24% chance pertaining to women whose pregnancies had all been lost. The importance of obstetrical history in predicting risk for SAB has been confirmed in other studies.[4,,2-,5] Such studies demonstrate that even a single SAB, unless there has been a subsequent successful pregnancy, increases the risk that the next pregnancy will terminate in another SAB. This blurs the distinction between "sporadic" and "recurrent" abortion and emphasizes, again, the importance of understanding risk factors for SAB as a step in understanding recurrent abortion.

RISK FACTORS FOR SPONTANEOUS OR RECURRENT ABORTION

Demographic Factors

Age

There is evidence for an increase in risk for SAB with increasing maternal age, especially in the subgroup of women who have not already experienced a pregnancy loss.12 Rates of SAB are less than 12% before age 25 and increase to 18o/o after age 39. Regarding paternal age, it has not been conclusively documented that the age of the father is a risk factor for SAB independent of maternal age. However, it is known that autosomal dominant disorders such as achondroplasia increase with paternal age[6] as well as trisomy 21 so that a paternal effect would not be surprising.

Racial/Ethnic Differences. Racial differences in the frequency of SAB have *not* been identified; but higher rates of SAB have been reported in some'[8] but not all ethnic subgroups[19] in which rates of consanguinity are high.

Genetic Factors

Cytogenetic Abnormalities

In a frequently cited case series in which aborted specimens were karyotyped, the frequency of abnormalities in consecutive specimens was 53.9% with the majority of abnormalities due to autosomal trisomies followed by 45X and triploidy.[20] The frequency *of* karyotypic abnormalities in abortuses from women with recurrent losses did not appear to be appreciably higher compared with an unselected series of abortion specimens. A gestational age of 9 weeks for lost pregnancies was most likely to be associated with an abnormality, although it is possible that losses related to a karyotypic abnormality at earlier gestational ages were underestimated because they failed to yield adequate tissue for study. The frequency *of* abortions associated with karyotypic abnormalities, however, decreased among specimens from losses at older gestational ages, especially after 16 *weeks*. Maternal age was found to predict the likelihood of a karyotypic abnormality, especially one associated with an autosornal trisomy. Despite the high

frequency of karyotypic abnormalities found in SAB specimens, chromosomal studies of couples with recurrent abortion reveal parental abnormalities in only 3—5% of couples, which is nevertheless about five times higher than in the general population.[21] The most common abnormality is a balanced translocation.

Other Genetic Abnormalities

Because karyotyping detects only gross chromosomal abnormalities, the contribution of genetic factors to the etiology of SAB may be higher than the estimated 50—60%. Single-gene mutations or polymorphisms are possibly responsible for Losses not recognized to have a genetic basis, but *the* scope of the problem is unknown. In the study by Kajii et al,[20] there was an excess of "male" abortuses among the specimens that were chromosomally normal, suggesting that lethal X-linked mutations may play a role. Indeed, women with glucose-6-phosphate dehydrogenase (G6PD) deficiency have a rate of SAB that is double the rate in a control group. Mutations predisposing to thrombophilia, including factor V Leiden mutations,[23] may be another example of a single gene associated with recurrent loss.

Structural Abnormalities of the Uterus

Women with congenital abnormalities of the uterus appear to have an increased risk for SAB or stillbirth. In particular, women with a unicornuate uterus are more likely to have an SAB.24 Women with a bicornuate or septate uterus may also have an increased risk for SAB.2[4] Mechanical problems, such as bifid or infantile uterus, can result in physical constraint, uterine distortion, and a subsequent reduction in blood supply to the fetus.[25] Submucosal and intramural uterine myomata may also contribute to mid-gestation abortion.[25]

Incompetent cervix is defined as asyrnptomatic dilation of the internal cervical canal leading to membrane rupture and premature delivery. It is associated with other congenital defects of the uterus, including those caused by fetal exposure to diethylstilbestrol (DES), and may result in midtrimester loss.26 A case-control study found that women exposed to DES in utero had a significantly smaller surface area of the endometrial cavity as compared with unexposed controls.[27] Wilcox et al[9] reported a twofold increase in risk of SAB in women with prenatal exposure to DES.

Endocrine Abnormalities

Thyroid Disorders

A small cohort study of hypothyroid women did not reveal an excess of SAB.[28] Nevertheless, one report suggested that hypothyroid women who had possibly been exposed to radioactive iodine from a nuclear power station had an increased risk for SAB.[29]

Diabetes

In a large collaborative study of diabetic and nondiabetic women enrolled prior to or early in pregnancy, insulin-dependent diabetes mellitus (IDDM) was not found to be an independent risk factor for SAB.[30] However, a subgroup of women with poor control of their diabetes, as indicated by elevated levels of glycosylated hemoglobin, had an increased rate of SAB of about 30% with each increase of 1 standard deviation above the normal range.

Luteal Phase Defect

Luteal phase defect (LPD), also called corpus Luteum defect or progesterone deficiency, is generally based on an endometrial biopsy that is "out of *phase"* by three or more days or a mid luteal progesterone level of less than 9—10 ng/mL. Case series of women with recurrent abortion describe LPD as a potential factor in 20 to 25% of cases.[23] A

meta-analysis of clinical trials of progesterone supplementation in women with LPD suggested that this might be efficacious to prevent SAB in a subsequent pregnancy; however, these were not randomized, double-blinded studies.[3]

High Luteinizing Hormone, Hyperandrogenism, and Hyperprolactinemia

A higher frequency of miscarriage among women with polycystic ovarian syndrome (PCOS) has been described.[32] An association between PCOS and RAB was found in another case-control study detecting ultrasonic evidence of PCOS in 820/o of women with a history of RAB.3~ In a subsequent prospective study,[34] ultrasound-confirmed PCOS was more frequent in women with RAB than in controls without a history of RAB.

A case-control study of women with otherwise unexplained RAB revealed significantly higher levels of prolactin and androstenedione during the follicular phase among cases compared with infertile women without a prior SAB having diagnoses of tubal factor or male infertility[35] Interestingly, women undergoing in vitro fertilization (IVF) who have higher luteinizing hormone (LH) levels on the day of hCG administration have a reduced rate of fertilization and cleavage, embryo viability, and clinical pregnancy.[35] Despite these observations, a controlled clinical trial of a regimen involving down-regulation with a gonadotropin-releasing hormone agonist, ovulation induction, and luteal phase progesterone failed to improve the term pregnancy rate significantly.[36]

Infection

Brucella is a well-known cause of abortion in farm animals. A report of pregnancy loss in women caring for farm animals suggested *the possibility of* an infectious etiology for SAB in humans.[38] However, more than eight decades after this observation was made, the precise role played by infection in SAB or RAB is still unclear. Infections with the capacity to colonize the female reproductive tract have been the subject of most investigations.

Chlamydia and Mycoplasma

A prospective study obtained cultures for chlamydia and two mycoplasma strains *(M. hominis* and *Urea plasma urealyticum)* from women at their first prenatal visit and failed to find an increased risk for SAB among women with any of the three infections.[39]

Bacterial Vaginosis (BV)

BV is an alteration of the normal vaginal bacterial flora associated with the loss of lactobacilli and overgrowth of anaerobic bacteria, including *Gardnerella vaginalis* and mycoplasma. It is diagnosed clinically as a thin, gray vaginal discharge that displays a high pH (>4.5), the presence *of* "clue" cells (vaginal epithelial cells with adherent bacteria), and a fishy odor when mixed with potassium hydroxide. A prospective study of women undergoing IVF suggested higher rates of SAB in women who had BV; however, the relevance of this observation to natural conception may be debated.

Genital Herpes

Studies suggest *an increased* risk I or SAB among women with genital herpes, especially when an initial (primary) infection occurs early in pregnancy.[41,42]

Other Health Conditions

A high rate of SAB has been observed in infertile women with endometniosis.[3] High levels of phenylalanine are thought to interfere with embryonic cell metabolism.[25] A significant increased risk of SAB has been documented among women with phenylketonuria, with the risk being greatest among women who remain untreated for their

condition. Severe nutritional deprivation can contribute to pregnancy loss at any stage of pregnancy.

Immunologic Factors

Autoimmunity

Antiphospholipid syndrome (APS) is an autoimmune disorder characterized by the presence of increased *levels of* antiphospholipid antibodies (e.g., anticardiolipin (ACAI antibodies or lupus anticoagulant [LAC]) and one or more features such as pregnancy loss, thrombosis, or thrombocytopenia. In a series of 242 untreated pregnancies in 65 women with LAC, 91% terminated in SAB or stillbirth.[43] In a series of women with two unexplained losses, 7% had LCA and 190o had ACA, proportions much higher than the control population.[44]

Alloimmunity

This term refers to immunologic differences between individuals of the same species (e.g., traits such as blood groups and histocompatibility antigens). A popular theory is that allogenic similarities between reproductive partners might lead to recurrent loss.[45] Treatment strategies aimed at immunizing the female partner with the male partner's white blood cells or with immunoglobins appear to be of questionable benefit.[46,47]

Environmental Factors

Smoking

Several studies have shown a positive association between maternal smoking and risk of SAB.[25~4~5]O Current smokers have on the order of 25—50~/~ increased risk of SAB compared with nonsmokers, and the risk increases with the number of cigarettes smoked per day.[4~52] One case-control study found an association between SAB and maternal exposure to environmental tobacco smoke (ETS) for at least 1 hour per day[53]; however, a prospective study found little evidence to support this association.[48] Most studies do not show a decline in male fertility associated with paternal smoking.[54]

Alcohol

An increase in the risk of SAB associated with alcohol intake has been reported in a number of studies,[25~48~55__57] the highest risk being associated with an intake of greater than 6 oz/day.[2,] Harlap and Shiono (1980) reported that moderate alcohol consumption increased the risk of second- but not first-trimester losses.[5,]

Caffeine

Consumption of caffeine is considered a weak risk factor for nonclinicallyS[8] and clinically recognized pregnancy losses.[52,,,63] A slight increase in risk for SAB has been linked to moderate (fewer than three cups or <300 mg per day) caffeine intake in the first trimester.[525~264] One case-control study comparing serum levels of a caffeine metabolite in women with and without SAB found higher levels among cases; however, this translated into risk only for very high levels (>300 mg/day).[64] Other studies on this topic have shown mixed results.[65] A potential bias is that women with a viable pregnancy are more likely to experience nausea and reduce their caffeine intake than women with a nonviable pregnancy, resulting in a spuriously overestimated relative risk.[66] However, a case-control study observed that among women experiencing nausea, there was still a significant increase in SAB risk associated with caffeine intake greater than 300 mg/day.[6,]

Trace Elements

Selenium (Se) is an essential cofactor for the antioxidant enzyme glutathione peroxidase. Although two studies found significant decreases in serum Se among women who miscarried as compared to women with normal pregnancies,[67,68] a larger case-control study did not confirm this association.[69] A review of the epidemiologic literature suggests that paternal exposure to lead or mercury may be associated with an increased risk of SAB.[7~] At levels below 50 pg%, lead does not appear to in-crease the risk of abortion.[2,] Maternal exposure to lead or mercury is not a determined risk factor for SAB.[7,] In a study investigating the impact of heavy metals on hormonal and immunological factors in women with a history of RAB, urinary mercury excretion was significantly associated with the number of amalgam tooth fillings among primary aborters and inversely correlated with progesterone levels.[7,]

Tap Water

A series of five retrospective studies conducted in one California county examined the risk of SAB associated with the source and amount of drinking water consumed during early pregnancy.[727,] Data from four of these studies were consistent with a 10—50% increased risk of SAB in women who drank tap water (~6 glasses per day) compared with women who did not drink tap water,[727,] whereas tap water consumption was not associated with SAB in a fifth study.[76] A larger prospective study[77] reported an even stronger association between tap water and SAB than what was previously reported in 1992. Trihalomethanes are common contaminants of chlorinated tap water[78]; high exposure to bromodichloromethane, in particular, was associated with a significant twofold increase in risk of SAB.

Other Environmental and Occupational Exposures

Studies have shown conflicting results with respect to anesthetic gases.[79,80] Two studies have refuted the hypothesis that exposure to video display terminals for more than 20 hours/week is related to SAB.[8182] One case-control study found an increased risk of SAB associated with standing for more than 8 hours/day at work.[8,] Work involving a large biomechanical load, specifically high peak pressure scores, showed significantly elevated odds for spontaneous abortion; this risk was most significant for work involving bending.[84] An elevated risk for spontaneous abortion was observed among women experiencing high levels of job stress.[8,] Taskinen et al'[6] reported significant associations between SAB and exposure to toluene, xylene, and formalin for a period of at least 3 days to a week. As with all teratogens, however, the magnitude of risk is directly related to the dose.[2,]

DIRECTIONS FOR FUTURE STUDY

We believe that our examination of the topic of recurrent abortion from an epidemiologic perspective leads to some conclusions that may affect the direction of both future clinical and epidemiologic studies. First, it seems clear that even a single pregnancy loss increases the risk for a subsequent abortion. This implies that there may be no clear distinction between risk factors for spontaneous abortion and risk factors for recurrent abortion. The latter must be understood in the context of risk factors for spontaneous abortion.

Second, any attempt to identify modifiable risk factors for SAB or RAB must deal with the fact that at least 50% of SABs are associated with cytogenetic abnormalities. Unless the study is designed to distinguish losses at least associated with gross chromosomal defects, researchers must be prepared to accept the fact that odds ratios for associations of interest will be biased toward the null. For example, it is unlikely that genital infections would affect the likelihood of a karyotypic abnormality in an embryo, thus studies based on karyotypically normal abortuses would be better able to distinguish the role of genital infection as a modifiable risk factor. Because of the high cost and difficulty of cytogenetic studies of abortion specimens, this is not an easy issue to resolve. However, it is likely that only a large and comprehensive study that includes cytologic examination of the aborted specimens will be successful in identifying risk factors for SAB or RAB. Alternatively, studies distinguishing normal from cytogenetically abnormal specimens are likely to show stronger associations with commonly identifi9d risk factors when compared with studies that neglect to make this distinction.

Finally, there is a clear need for clinical trials of therapy for RAB that meet epidemiologic standards including

randomization, double blinded-ness (whenever possible), and placebo control. As mentioned earlier, recurrent abortion is arbitrarily defined as three consecutive pregnancy losses. Allowing women with two consecutive losses to be included in case series of women with recurrent abortion would increase sample sizes and facilitate the design of therapeutic clinical trials that include a placebo arm. Given the high emotional and financial costs of some of the existing therapies, patients and clinicians should require proof of efficacy from well-designed clinical trials.

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