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Invited Editorial: Influence of MHC and MHC-linked Genes on Reproduction

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Recurrent pregnancy loss of unknown etiology is both an "experiment of nature" that may provide fundamental insights into the processes of embryogenesis and implantation and a frustrating and emotionally charged clinical problem. Under the best of circumstances, the reproductive process in humans is quite inefficient: approximately 75% of fertilized ova are lost (Gill, in press). Most of these losses may be accounted for by chromosomal abnormalities in the conceptus, infection, hormonal imbalances, and anatomical abnormalities of the maternal genito-urinary system. Even when all of these factors are taken into consideration, there still remains approximately 0.4% of couples who experience repeated pregnancy losses without any apparent cause: recurrent spontaneous abortion is defined as three or more pregnancy losses in a woman with the same partner. Sometimes these couples are further categorized as either primary aborters, if they have no live births, or secondary aborters, if they have one or more live births. The etiological significance and clinical utility of this subclassification are not yet clear.

Couples who have recurrent spontaneous abortions may have other types of reproductive failures (Coulam et al. 1989, 1991), and there is an increased prevalence both of cancer in these women (Ho et al. 1989; Gill, in press) and of congenital anomalies in any children whom they may eventually have (Gill, in press). In addition, there is an increased prevalence of recurrent spontaneous abortions, cancer, and congenital anomalies in the first, second, and third-degree relatives of these couples (Ho et al. 1991*b*).

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Recurrent spontaneous abortion may be more a syndrome than a disease with a single etiology. Nonetheless, the same general mechanism may underlie the different manifestations of this disease, and those mechanisms that have been postulated to play a major role are either immunologic or genetic. It is important to elucidate the basic mechanism of recurrent spontaneous abortion not only to understand its etiology but also to make the proper therapeutic decisions for those suffering from it. If the mechanism is immunological, manipulation of the immune system may be therapeutically efficacious. If it is genetic, counseling would be the most appropriate choice of treatment at this stage of our knowledge, and active therapies that may not be specifically effective could be avoided.

One major line of conceptual thinking about the implantation process grew out of the field of tissue transplantation, and it was cast in terms of the "survival of the fetal allograft" (Medawar 1953; Billingham 1964). A large number of experiments have been directed along basically transplantation lines: the observation of local sensitization of the uterus by skin grafts (Beer and Billingham 1974), the production of "blocking" antibodies during pregnancy (Takeuchi 1990), and the association of cytokine production with the implantation process (Clark et al. 1991; Guilbert et al. 1991). They fit the concept of an immunological mechanism's being essential to the process of implantation and, by inference, its absence being detrimental. The early observations in the literature that showed an increased sharing of HLA antigens in couples having recurrent spontaneous abortions (Gill 1983) were considered, according to this school of thought, as being consistent with an immunological mechanism. A series of experiment using matings of CBA ($H\text{-}2^k$) × DBA/2 ($H\text{-}2^d$) mice (Clark et al. 1980; Chaouat et al. 1983), which show spontaneous fetal losses, led to the conclusion that immunization with lymphocytes from the BALB/c ($H\text{-}2^d$) strain but not from the DBA/2 strain prevented these losses. The

mechanism underlying this observation has not been elucidated. The production of blocking antibodies or the elicitation of specific cytokines was postulated to be involved; immunogenetic analysis of the results failed to provide an explanation (Gill 1990). Nonetheless, these experiments were taken to support the immunological hypothesis of implantation.

Based on the immunological interpretation of these clinical and experimental observations, a variety of attempts to treat recurrent spontaneous abortion in humans by immunization of the woman, either with her husband's lymphocytes or with third-party lymphocytes, were undertaken. The results have been controversial, and, although there has been much discussion of the subject, relatively few systematic clinical studies testing the efficacy of this modality of treatment have been reported. One prospective study (Mowbray et al. 1985) reported that immunization improved reproductive capacity in recurrent spontaneous aborters, but two other studies (Cauchi et al. 1991; Ho et al. 1991*a*) failed to show any effect of this treatment. An analysis of data from many sources—including many small studies and unpublished results—argues that under certain conditions immunization of recurrent aborters improves reproductive capacity (Clark and Daya 1991). Nonetheless, in my opinion, this mode of therapy has not yet been shown to be physiologically useful—the psychological aspects have yet to be studied adequately.

The alternative explanation for recurrent spontaneous abortion, i.e., that it has a genetic basis, is predicated on the large number of observations showing that lethal and semilethal genes are present in the mammalian genome (Gill et al. 1983; Strickberger 1985). From this point of view, the observation that there is increased HLA sharing in couples experiencing recurrent fetal losses (Gill 1983; Reznikoff-Etievant et al. 1988; Ho et al. 1990) is interpreted as the antigen sharing's being just a marker for the sharing of HLA-linked genetic defects, either recessive lethal genes or deletions of genes necessary for normal growth and development, that combine in the embryo to cause its demise (Gill 1984, 1987, and in press). In addition, these genes may also be involved in the pathogenesis of congenital anomalies and in the increased susceptibility to cancer (Gill 1984, and in press).

Evidence for this hypothesis comes both from experimental studies in rats and from clinical observations in humans. Strains of rats having a deletion (*grc*$^-$) (Vardimon et al., in press) in the major histocompatibility complex (MHC)–linked growth and reproduction complex (Kunz et al. 1980) have the following phenotypic characteristics when homozygous: small body size, male sterility, and reduced female fertility (all three of which are developmental defects); perinatal loss of approximately 25% of their offspring; and increased susceptibility to cancer induced by chemical carcinogens, possibly because of the loss of genes that are, among other things, tumor-suppressor genes (Melhem et al. 1989, 1991). The number of fetal losses in the *grc*$^-$ strains is increased by interaction with non-MHC–linked, heterozygous recessive lethal genes (lethal epistatic interaction) (Schaid et al. 1982). Similar MHC-linked genes are present in other species of animals as well (Gill, in press), and they have various effects on reproduction; there is some evidence that they also exist in humans (Degos et al. 1974; Alper et al. 1986).

Studies in humans have shown that the increased HLA sharing in couples experiencing recurrent spontaneous abortion can occur at all loci and is not restricted to one specific HLA locus, although the sharing at HLA-DR appears to occur at a higher frequency than the sharing at the HLA-A and HLA-B loci (Gill et al. 1983; Schacter et al. 1984; Weitkamp and Schacter 1985; Coulam et al. 1987; Ho et al. 1990). Lethal epistatic interaction between MHC-linked genes (or, possibly, deletions) and non-MHC genes also occurs in humans (Weitkamp and Schacter 1985). The strongest correlation between HLA sharing and either recurrent spontaneous abortion (Ho et al. 1990) or gestational trophoblastic tumors (Ho et al. 1989) is the sharing of three or more of the HLA-A, -B, -DR, or -DQ loci in couples with either disease. This observation strongly suggests that HLA sharing is just a marker for the sharing of HLA-linked genes or deletions, such as those in the *grc*$^-$ rats, and that the sharing of HLA antigens *per se* is not the pathogenic mechanism. Statistical models applied to population data on HLA sharing, maintenance of HLA polymorphism, and recurrent spontaneous abortion (Hedrick 1988; Hedrick and Thomson 1988) also support the genetic hypothesis for recurrent spontaneous abortion.

Three pieces of collateral evidence further support the genetic hypothesis for recurrent spontaneous abortion. First, there is an increased prevalence of recurrent spontaneous abortion, congenital anomalies, and cancer in the first-, second-, and third-degree relatives of the couples having recurrent spontaneous abortions (Ho et al. 1991*b*). This finding suggests that the genes, or genetic defects, involved in the genesis of these diseases are segregating at a higher frequency in the rela-

tives of aborters than in the general population. Second, pedigree analyses of a number of families have shown that recurrent spontaneous abortion can occur in three generations of the same family (Mowbray et al. 1991). Third, women who have recurrent spontaneous abortions with one partner often do not have them with another partner (Coulam et al. 1986).

The paper by Ober and her co-workers in this issue of the *Journal* (Ober et al. 1991) continues the work in their long-standing interest in the reproductive patterns among the Hutterites, who are an Anabaptist population that is highly inbred, has large families (contraception is proscribed), and has extensively documented pedigrees. The knowledge derived from this body of work provides some important insights into the mechanism of reproductive failure, and it is consistent with the genetic hypothesis about the pathogenesis of recurrent spontaneous abortion and related disorders.

Their early studies showed that the median intervals between births were longer among couples who shared more than one HLA antigen (Ober et al. 1983). These longer intervals were associated with an increased rate of spontaneous abortion among couples who shared HLA-DR antigens (27%), compared with couples who shared only HLA-A or HLA-B antigens (9%) or with couples who shared no HLA antigens (12%) (Ober et al. 1985). They interpreted this finding as suggesting a potentially important role for undefined HLA-linked genes—most likely in the DR region (Ober et al. 1985, 1988)—in normal pregnancy. Significant differences in sex ratios, minor anomalies, and size were also observed in HLA-DR-compatible offspring compared with HLA-DR-incompatible offspring (Ober et al. 1987). Their present paper confirms their previous conclusions in a prospective, 5-year study of fecundability and of the rate of fetal losses in this Hutterite population. It emphasizes again the role of HLA-DR sharing in these measures of reproductive capacity and presents evidence for a role for HLA-B also. These observations, as well as that of the importance of the DQ locus (Ho et al. 1990), suggest that the critical HLA-complex region that affects reproduction is delineated by the DQ-DR-B loci and that the loci encoding the HLA antigens themselves are not the critical factors.

The weight of evidence in the literature, in my opinion, supports the genetic hypothesis for the etiology of recurrent spontaneous abortion and related reproductive defects. These genetic factors also influence some aspects of the susceptibility to cancer and are consistent with the relationship between embryogenesis and carcinogenesis, first postulated by Virchow (1860). The implications for the basic mechanisms of embryogenesis, implantation, and development are that genetic defects more subtle than chromosomal abnormalities are critical in determining the fate of the embryo and that at least some of the genetic factors responsible are located in the HLA-DQ-DR-B region of the MHC. If these genetic defects in the human are like those in the rat, they entail the loss of genes that are necessary for normal development, some of which may be tumor-suppressor genes. Thus, the chromosomal segment that includes genes critical for the recognition of self-nonself (MHC) also includes genes critical for the control of normal growth and development (MHC linked). This constellation of genes provides an even more cogent rationale for the evolutionary conservation of the genomic region carrying the MHC and its linked genes.

As a working hypothesis, we may look at different types of growth failure as involving, at least in some circumstances, various levels of defects in MHC-linked genes. The most severe defects would lead to the death of the embryo (recurrent spontaneous abortion); less severe defects would lead to congenital abnormalities; and the least severe defects would lead to a predisposition to cancer, possibly by providing the "first hit" (Knudson 1985) in the carcinogenic process.

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