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Physiopathological determinants of human infertility

The ESHRE Capri Workshop Group*

Successful management of infertility involves the use of appropriate diagnostic tests and treatments, and knowledge of prognostic factors such as age of the female partner and duration of infertility. The assessment of infertility may reveal disorders that cause morbidity in the normally fertile population, which might or might not contribute to the infertility. Endocrine dysfunction is a significant cause of infertility due to amenorrhoea and dysfunctional uterine bleeding, and hirsutism is a familiar problem in the normal population. Some genetic abnormalities cause infertility in males and females and create implications for the progeny of infertile couples. Sexually transmitted diseases cause illness and healthcare expenditure among young adults, and can also result in tubal infertility. Congenital disorders of the uterus and fibromyomas are not rare and they are frequently asymptomatic, but in some cases uterine anomalies may contribute to infertility and pregnancy loss. Autoimmunity underlies some common diseases, especially among females, and it can also be associated with pregnancy loss. When such widely distributed factors are identified during the diagnostic assessment of subfertile couples, it is important to distinguish between a coincidental association and a specific relationship to the infertility.

Key words: anovulation/assisted procreation/genetic abnormalities/infertility/sexually transmitted disease

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Introduction

Appropriate management of infertility depends not only on sensible selection of diagnostic tests and treatments, but also on understanding the way in which non-medical factors may affect the development of subfertility and the likelihood of conception among couples with subfertility. Age, particularly that of the female partner, has a bearing on time to conception in all couples, and particularly in those who have deferred conception until the woman is in her mid thirties. Also, the duration of infertility contributes meaningful information to the likelihood of future fertility because, in many cases, in the absence of a specific diagnosis the length of exposure without conception reflects the presence of unknown genetic and pathological factors; the longer the duration of infertility, the more powerful the underlying factors. There are a growing number of known genetic factors which may indirectly or directly cause infertility in males and females. These factors need to be considered together with types of treatment in a coherent context because of the implications for future generations. There are also diagnostic categories contributing to infertility which have broader implications in the normal population. Endocrine dysfunction is a significant cause of morbidity and provides an important group of defects contributing to the diagnostic category of ovulatory infertility. Hirsutism is a significant clinical problem in the normal population, while in some cases its endocrine causes may contribute to infertility. Sexually transmitted diseases are a significant cause of illness and healthcare expenditure among young adults, and the aftermath in a number of cases may be tubal infertility. Congenital anomalies of the uterus and fibromyomas may be asymptomatic, but in some cases may be implicated in delayed conception and pregnancy loss.

Autoimmunity, which leads to several common diseases—especially among females—can have a bearing on time to conception and pregnancy loss. This review addresses the

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significance of these pathophysiological determinants of infertility in order to provide a context for the diagnosis and treatment of infertility. Other factors which could not be included were weight gain and loss, nutritional disorders, cigarette smoking and substance abuse. Although the factors discussed may contribute to the development of subfertility and to the likelihood of conception among couples with subfertility, they are commonly found also in fertile people and care should be taken determine the specific relationship to infertility in each couple.

Age limits of normal and assisted procreation

The age of the infertile partners is an important factor to be considered in the provision of infertility services, in the clinical management of infertility and in the design of clinical trials. The number of women seeking infertility services is growing because the number of women aged 35 to 45 years is steadily increasing. For example, in the United States it is expected to increase from 13 to 18.5 million between 1980 and 2010 (Fonteyn and Isada, 1988). In clinical management and trial design, the prognostic effect of age affects decisions and plans. Here, the analysis of the effect of age on fertility will focus on women, since much less is known on the effect of age on the male partner. The effect of female ageing on fertility and pregnancy has been extensively reviewed (Maroulis, 1991).

The effect of age on reproduction depends on three components:

1. Intrinsic ageing.
2. The cumulative effect of various experiences on the reproductive system over the years.
3. Environmental, social, and financial conditions.

The effect of age on reproductive potential will be analysed by discussing fertility rates in non-contracepting populations and in treatment cycles. Overall fertility rate is defined using the number of live births in a specified time as numerator, divided by the total population of women of reproductive age (15–44 years). Age-specific fertility rates can be estimated for different age groups within the reproductive age range. Relative fertility is defined as the ratio of the fertility rate in one age group divided by the fertility rate in a reference (usually younger) group.

Fertility rates in untreated populations

Limited data exist on fertility rates in non-contracepting populations (17th century studies in Europe, American Hutterite women in the 1940s and contemporary women; see below). Fertility rates remain relatively stable until about the age of 30–32 years, when a decline begins; this steepens after the age of 40 years. Significant differences in fertility rate are observed between populations within the same age groups, although the rate of fertility decline has been remarkably similar in eleven historic communities from 1600 to 1950 (Henry, 1961), in 1225 Hutterite women from 1946 to 1950 (Eaton and Mayer, 1953), in 45 developing nations from 1960 to 1975 (Robinson, 1987), and in 4104 women in a developed nation (Oxford Family Planning Study; FPA) during the 1970s and 1980s (Howe *et al*., 1985) (Figure 1).

Due to the use of contraception in contracepting populations, the rate of decline is more rapid. US surveys from 1955 and 1981

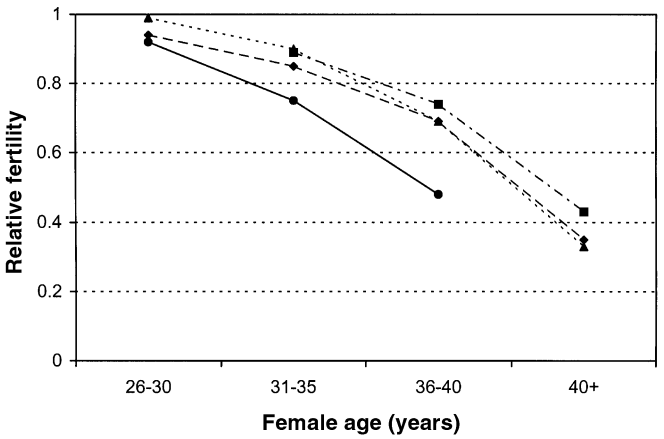

Figure 1. The decline of fertility with age in non-contracepting populations. ◆, 11 communities from 1600 to 1950 (Henry, 1961); ■, Hutterite women from 1946 to 1950 (Eaton and Mayer, 1953); ▲, 45 developing nations from 1960 to 1975 (Robinson, 1987); ●, Oxford Family Planning Study (Howe *et al*., 1985). Reference group: younger women.

indicate: (i) a progressive decrease in fertility rate within the same age groups; and (ii) a more rapid relative decrease in fertility with age when compared with natural, non-contracepting groups, this being a probable consequence of widespread use of contraception (National Center for Health Statistics, 1981). In population studies where late marriage was common and there was no evidence of fertility control, women aged over 40 years are not, as often portrayed, hopelessly infertile, but can achieve a pregnancy in up to 48% of cases (Maroulis, 1991).

The role of the male partner's age has not been well studied. Data obtained from a study analysing genealogical data (mid-19th century) showed that although wives had a decline in fertility after the age of 35 years, in men the decline began at age 40 years and, by the age of 64 years the fertility rate had fallen to 36% relative to men aged between 20 and 40 years (Mineau and Trussel, 1982).

Fertility rates in treatment cycles

A substantial amount of data exists regarding the effect of age on fertility rates in donor insemination (DI) and IVF cycles. DI cycles provide a good indicator of the fecundability (the probability of achieving a pregnancy within one menstrual cycle) of normal women in whom at least tubal and male factors can be excluded. The cumulative pregnancy and monthly fecundity (the ability to achieve a live birth from one cycle's exposure to the risk of pregnancy) rates in more than 2000 women who received DI decreased with age, especially after age 35 years (Fédération CECOS *et al*., 1982). In 37 000 conventional IVF cycles, reviewed by the British Human Fertilisation and Embryology Authority (HFEA), the overall live birth rate per cycle of treatment initiated was 13.9%. The highest live birth rates were in the age group 25–30 years; younger women had lower rates and there was a sharp decline in older women (Templeton *et al*., 1996). The influence of women's age is also evident in ICSI cycles, with a gradual but significant decrease in viable pregnancy rates occurring with advancing age. No viable pregnancies ensued in women from age 45 years onwards (Grimbizis *et al*., 1998a).

Impact of ageing on pregnancy outcome

Publications on the influence of ageing on pregnancy outcome provide conflicting results. Different factors of pregnancy outcome have been evaluated including low birth weight, premature labour, preterm delivery, premature rupture of membranes, fetal death and stillbirth. When corrected for medical complications which increase with age, delaying childbirth poses little if any increased risk for impaired neonatal outcome among patients receiving excellent medical attention. This may not hold true for women from all socio-economic strata, but even in disadvantaged groups there is no evidence of an increased risk due to age alone (Berkowitz *et al*., 1990).

Impact of ageing on spontaneous abortion

Large demographic studies have shown that the incidence of clinically recognizable spontaneous abortion in mothers aged over 40 years is of the order of 25–30%, but there are indications that most early pregnancy losses are occult, so the actual rate may be higher. A number of abortions go unrecognized, and to what extent this varies with age—either in natural pregnancies or following infertility treatment—cannot be accurately estimated. Despite these problems, many studies show an increase in abortion rate with age: 7–15% in women aged 20–29 years, and 21–46% in women aged 40 years and over (Jansen, 1982; Hansen, 1986).

Few data are available regarding the relationship between paternal age and spontaneous abortion; some suggest an increased risk of Down's syndrome with paternal age, while others do not reveal a paternal age effect (Erickson, 1978; Magenis and Chamberlain, 1981).

The impact of the duration of infertility on clinical management

Younger women, previously pregnant and with a short duration of infertility are more likely to become pregnant without treatment, as well as become pregnant with treatment. The decision as to when it is appropriate to treat is therefore crucially dependent on the size of these factors and how they weigh with other factors in determining outcome.

Three important studies have attempted to establish the relative importance of duration of infertility in predicting the likelihood of live birth in infertile couples, without treatment. The first involved 996 couples in Utrecht (Eimers *et al*., 1994). Duration of infertility had an adjusted fecundity rate of 0.89 (CI 0.82–0.96) per year, with very low likelihood of pregnancy after 7 years. This study was confined to women with no cycle disturbance or tubal disease. The second study in 11 Canadian Fertility Centres involved 2198 couples among whom 263 live births had occurred (Collins *et al*., 1995). Women with a duration of infertility of 36 months or less had an associated likelihood of live birth of 1.7 (95% CI 1.1–2.5). In comparison, the figure for previous pregnancy was 1.8 (CI 1.2–2.7) and for age less than 30 years it was 1.5 (CI 1.1–2.2). This study included all infertile couples, regardless of diagnosis, and it was possible to apply relative hazards analysis to certain diagnostic groups, including male problems (0.5, CI 0.3–0.8), endometriosis (0.4, CI 0.2–0.9) and tubal defects (0.5, CI 0.4–0.6). In a third, population-based study of 762 couples in Walcheren (The Netherlands), there were 201 live births and 342 couples remained untreated (Snick *et al*., 1997). The adjusted relative likelihood of a live birth when the duration of infertility was less than 24 months was 1.5 (CI 1.1–2.1) and was similar whether an abnormal post-coital test (PCT) was included in the prediction model, or not. An important difference between the latter studies was a much shorter median duration of infertility in the Dutch study (16 versus 36 months). The following estimates of adjusted relative hazard for live birth per year of infertility duration were provided: 0.87 (CI 0.83–0.90) and 0.89 (CI 0.80–0.99) in the Canadian and Walcheren studies respectively.

Table I. Duration of infertility and IVF success

Duration of infertility (years)	Live birth rate per treatment cycle
1–3	15 (15–16)
7–9	13 (12–14)
>12	9 (7–10)

Turning to the impact of duration of infertility on the outcome of treatment, an analysis of the UK database of IVF cycles indicated that duration of infertility had a significant impact on the live birth rate, when adjusted for women's age (Templeton *et al*., 1996). Subsequent analysis including 44 236 cycles (Templeton and Morris, 1998) indicated that each year of infertility diminished the odds of a birth by 0.98 (CI 0.98–0.99), as well as the odds of a multiple birth (0.98, CI 0.97–0.99) (Table I).

A comparison of expected spontaneous pregnancy rates and IVF pregnancy rates indicates that in a number of clinical situations a policy of expectant management would be prudent, particularly if IVF treatment will continue to carry a significant risk of twins and triplet pregnancy.

Endocrine dysfunction as a determinant of human infertility

Reproduction is controlled by the endocrine system and is subject to influences including season, photoperiod and nutrition. Because the energy expenditure of pregnancy and lactation is considerable, control is usually exerted through the female rather than the male. It is not surprising therefore that disorders of the endocrine system are a much more common cause of infertility in women than in men.

Natural periods of infertility

Physiological periods of infertility occur naturally pre-pubertally, during pregnancy and lactation, and post-menopausally. At the extremes of reproductive life there is a period of relative subfertility when anovulatory cycles become more common (Treolar *et al*., 1967).

Pathological causes of infertility

The proportion of endocrine causes of infertility varies from country to country. In Western Europe and North America, where tubal disease is relatively uncommon, endocrine dysfunction can

be identified in about 10–20% of women presenting with infertility (The ESHRE Capri Workshop Group, 2000). The primary defect may lie in any one component of the hypothalamic–hypophyseal–ovarian axis leading to anovulation. The most useful classification is a functional one in which the women can be divided into one of five categories (Baird, 1997): (i) hypergonadotrophic hypogonadism; (ii) hypogonadotrophic hypogonadism; (iii) hyperprolactinaemia; (v) normogonadotrophic anovulation; and (v) miscellaneous.

Successful identification of the cause is important because it dictates the choice of treatment.

Hypergonadotrophic hypogonadism

Hypergonadotrophic hypogonadism (Baird, 2001) is characterized by raised levels of FSH and LH indicating ovarian failure. Almost always, this is due to depletion of oocytes, although rarely it may represent resistance to gonadotrophins due to a receptor defect.

Hypogonadotrophic hypogonadism

The causes of failure of secretion of FSH and LH are multiple, but by far the most common in Western societies is weight loss. Anovulation can occur with modest losses (around 10% body weight) and is due to decreased secretion of GnRH by the hypothalamus. The exact mechanism is not fully understood, but it is probably similar to those mechanisms which protect female animals from the consequences of pregnancy when food is in short supply. Treatment with pulsatile GnRH is highly successful (Filicori *et al*., 2001).

Hyperprolactinaemia

Hyperprolactinaemia is associated with reduced secretion of gonadotrophins and hence anovulation. Minor degrees may not prevent ovulation but may cause an inadequate luteal phase such as occurs commonly during the establishment of cycles at puberty or after lactation. A pituitary adenoma can be identified in almost 50% of women with hyperprolactinaemia.

Normogonadotrophic anovulation

This group represents about 50% of women with an endocrine cause of infertility, and includes women with polycystic ovary syndrome (PCOS). The aetiology of this condition is unknown, but it has a genetic component which includes mutations in genes involved with metabolic homeostasis (Franks, 1995).

Miscellaneous

Various other endocrine-based conditions exist, one example being luteinized unruptured follicle (LUF) syndrome. The endocrine basis for this condition is not clear. Prostaglandins are involved in the mechanism of follicle rupture, and many women with LUF syndrome have a history of ingestion of non-steroidal anti-inflammatory agents.

Endocrine causes of male infertility

Endocrine causes of male infertility are relatively rare (Comhaire *et al*., 1987), although there is no doubt that infertility can occur in association with a number of defects in the endocrine system (Nieschlag and Behre, 1992; de Kretser and Baker, 1995). Decreased secretion of gonadotrophins occurs in association with Kallman's syndrome, and pituitary disease with or without hyperprolactinaemia results in a hypogonadal state (Nieschlag and Behre, 1992). Because of the reduced levels of testosterone sexual function is reduced or absent, as well as impairment of spermatogenesis (Santen, 1999). However, the functional causes of hypogonadotrophism seen in women are relatively uncommon in men except in association with severe nutritional deprivation.

Exogenous anabolic and/or androgenic steroids such as testosterone enanthate and mesterolone cause male infertility because they suppress gonadotrophins and hence spermatogenesis (Schänzer, 1998). In this case there are no clinical signs of hypogonadism except a reduction in the size of the testes.

Infertility in hyperandrogenic women

Ovarian causes of hyperandrogenism

These are of particular concern to the infertility specialist because the most common underlying condition is PCOS. The diagnosis is established from the history (symptoms usually date from menarche or shortly thereafter), the common finding of obesity, together with the characteristic ovarian ultrasound and endocrine features. Associated infertility is usually caused by a reduced rate of ovulation, so management is essentially focused on increasing the frequency of ovulation. Traditionally, treatment has involved anti-oestrogen medication which, if unsuccessful, is followed by gonadotrophin stimulation or ovarian diathermy. The latter induces ovulation in 65–85% of clomiphene-resistant patients. IVF is an attractive form of infertility treatment in patients in whom the ovarian response to gonadotrophin therapy is excessive and difficult to control, and in whom ovarian diathermy is unsuitable because of associated obesity.

During the past decade it has been increasingly recognized that many women with PCOS, and particularly those with hirsutism and oligomenorrhoea, are resistant to the glucose-lowering effect of insulin (Dunaif, 1997). The cause of this insulin resistance is multifactorial: in part genetic, in part the result of endocrine changes (the increase of growth hormone secretion at puberty), and in part the consequence of obesity. Irrespective of the mechanism(s), the consequence is the compensatory hypersecretion of insulin. Prolonged exposure to high levels of insulin results in the development of a metabolic syndrome of adverse cardiovascular risk factors (Syndrome X); it is also implicated in deteriorating ovarian function, as indicated by the onset of menstrual dysrhythmia and hyperandrogenism (Conway and Jacobs, 1993). Treatment is therefore focused on reducing insulin drive. Much emphasis is placed on changes to lifestyle, such as increasing energy expenditure by exercising more and reducing energy intake by eating less. Reducing body weight by as little as 5% may lead to the resumption of regular ovulatory menstrual cycles (Clark *et al*., 1995). Given the degree of obesity of many of these patients, the absolute weight loss required may be considerable, however. Interest has therefore turned to medication as a way of increasing the body's sensitivity to the glucose-lowering effect of insulin.

Three types of insulin-sensitizing agents have been used. The first and most widely studied is metformin, a biguanide used for many years in the treatment of type 2 diabetes. In patients with diabetes, this drug reduces hepatic glucose output (it lowers the fasting glucose concentration), increases glucose uptake by

muscle (it enhances extrasplanchnic insulin sensitivity) and reduces adipocyte lipolysis (Morin-Papunen *et al*., 2000). In women with PCOS, in whom circulating insulin levels are lowered by treatment, benefit derives predominantly from the latter two mechanisms. In some studies, a weight loss of ~2 kg over 6 months treatment was recorded. Most of the literature related to the use of metformin in PCOS is based on using a dose of 1.5 g per day, although 2.0–2.5 g per day may be more efficacious (Nestler, 2001). Only a handful of randomized trials has been published, but the consensus today is that, in PCOS, the main indication for treatment with metformin is induction of ovulation in overweight insulin-resistant women with anovulatory infertility (Norman *et al*., 2001). While total serum testosterone levels fall during treatment, little or no improvement in hirsutism has been recorded. Metformin is considered safe in pregnancy (Glueck *et al*., 2001; Iuorno and Nestler, 2001).

Troglitazone (Mitwally *et al*., 1999) and D-chiro-inositol (Nestler *et al*., 1999) have also been shown to correct hyperinsulinism in PCOS. The former drug has been withdrawn because of serious adverse effects on the liver; further studies of the latter are awaited. Since the thiazolidinediones presently available (rosi- and pioglitazone) have been shown in animal studies to retard fetal development, this class of drugs is unlikely to find a place in the management of anovulatory women with PCOS.

Adrenal causes of hyperandrogenism

These are less common than ovarian causes, but may have a significant impact on infertility and its management. The most important is 21-hydroxylase deficiency, which may either be congenital or of later onset. The former condition is the more severe, and may cause masculinization alone (simple virilizing) or in association with mineralocorticoid deficiency (salt wasting). Diagnosis is established by measurement of steroid precursors of the deficient enzyme, supplemented by molecular genetic characterization. Treatment is by glucocorticoid suppression of adrenal oversecretion of precursors together with replacement of cortisol and mineralocorticoid, as appropriate. Treatment is traditionally monitored by measurements of 17-hydroxyprogesterone and testosterone concentrations in serum.

It is important for the infertility specialist to realize that most women with 21-hydroxylase deficiency also have polycystic ovaries (Hague *et al*., 1990) and that glucocorticoid therapy causes insulin resistance. It is also important to recognize that, in patients with 21-hydroxylase deficiency, anovulation may be caused by overproduction of progesterone rather than of androgens (Holmes-Walker *et al*., 1995). Adrenal secretion of progesterone may occur as a result of poor therapeutic control; one such patient has been described in whom treatment by bilateral adrenalectomy led to the development of regular ovulatory menstrual cycles and pregnancy (Holmes-Walker *et al*., 1995). Finally, the impact of these women's life-time exposure to excessive amounts of androgen must be constantly respected.

Genetic determinants of male infertility

The genetic determinants of male fertility can be defined as the genetic conditions necessary to create a healthy male individual able to impregnate a female individual through intercourse. To put the known genetic causes of male infertility into context, it is first necessary to summarize 'normal' male sex differentiation and spermatogenesis. Normal male sex differentiation involves a correct genetic sex, which means that an X-bearing oocyte should be fertilized by a Y-bearing spermatozoon. This happens by chance, but once an XY-zygote exists and develops further, sex differentiation occurs in a well-timed sequential manner. The correct male gonadal sex, which means the development of testes from the undifferentiated gonads, needs at least the presence of SRY (sex-determining region of the Y) on Yp11.3 (Sinclair *et al*., 1990). Without SRY no testes will develop, but SRY is not sufficient for correct sex differentiation. A number of other genes on autosomes as well as on the X chromosome, which modulate or interfere with the SRY-protein action, are known today—others are as yet unknown. The correct phenotypic sex will depend on the action of androgens with androgen receptors as well as on the action of gonadotrophins and GnRH (Bogan and Page, 1994; Ramkissoon and Goodfellow, 1996; McElreavey and Fellous, 1997; Roberts *et al*., 1999; Veitia *et al*., 2001). Linked to all these well-defined developmental steps are spermatogenesis, spermiogenesis, ejaculation and the physical, mental and psychological abilities leading to the correct functional sex.

The more classical classification of the genetic determinants of male infertility is based on the presence or absence of chromosomal aberrations or monogenetic defects.

Chromosomal aberrations leading to male infertility or subfertility

These are often discovered because of reproductive problems:
(i) Numerical sex chromosomal aberrations such as 47XXY; 46XY/47XXY; 45X/46XY; 47XYY; and variants are the results of meiotic non-disjunctions during gametogenesis or, in case of mosaics, of mitotic non-disjunctions in the early embryo. They appear *de novo* and can be diagnosed by conventional karyotyping or fluorescence in-situ hybridization (FISH). Usually, these patients present with a male phenotype but have non-obstructive azoospermia or oligoasthenoteratozoospermia (OAT).
(ii) 46XX sex-reversal males are usually the result of unequal crossing over between X-Y bivalents near the pseudoautosomal regions during male meiosis. The phenotype is Klinefelter-like presenting with non-obstructive azoospermia. This aberration appears *de novo*. The conventional karyotype will display the female karyotype, but FISH and other molecular technology are necessary to establish the correct diagnosis.
(iii) Structural Y-chromosomal anomalies such as 46X, der(Y) in non-mosaic configurations or translocations between the Y chromosome and an autosome can lead to sex reversal, azoospermia or OAT according to the region of the Y-chromosome which is involved or deleted. They are usually *de novo*, and karyotyping completed with FISH and molecular studies will allow a correct diagnosis.
(iv) Robertsonian translocations and reciprocal balanced translocations between autosomes interfere with normal meiosis and can thus lead to OAT in men, although this is unpredictable and not yet understood. These anomalies can appear *de novo*, but are often inherited from one of the parents. Conventional karyotyping will in most instances disclose the anomaly. FISH technology is necessary to determine the breakpoint, but possible cryptic translocations could be missed unless subtelomeric FISH probes

Table II. Incidence of chromosomal abnormalities (%) in infertile oligospermic and azoospermic males compared with newborns. (Data derived from Van Assche *et al*., 1996.)

Chromosomal abnormality	Category			
	Infertile males ($n = 7876$)	Oligozoospermia ($n = 1701$)	Azoospermia ($n = 1151$)	Newborns ($n = 94\,465$)
Autosomes	1.3	3.0	1.1	0.25
Sex chromosomes	3.8	1.6	12.6	0.14
Total	5.1	4.6	13.7	0.39

are used. The introduction of FISH technology has allowed the study of segregation of involved chromosomes in sperm.
(v) Yq11 microdeletions are usually not seen with a conventional karyotype or with FISH. In this case, molecular techniques must be used to find a deletion and to characterize the extent of the deletion or the genes involved.

As the sperm count decreases, then the average number of chromosomal aberrations increases. Also, more numerical sex chromosomal aberrations occur with azoospermia and more structural chromosomal aberrations occur with OAT (Table II) (Van Assche *et al*., 1996).

Yq11 deletions are more frequent in azoospermia and less frequent in OAT. In men with azoospermia, the extent of the deletion (AZFa,b,c) may help to predict whether spermatozoa will be found at testes biopsy.

Known genes in the AZF region such as RBMY, DAZ, CDY, PRY, USP9Y, DBY and UTY seem to play a role in spermatogenesis by regulating gene expression and RNA translation (McElreavey and Krausz, 1999).

Monogenic defects

These play a role in male infertility, and often cause other associated problems:
(i) Monogenic defects causing abnormal sex differentiation and abnormal spermatogenesis:

- persistent Müllerian duct syndrome due to antimüllerian hormone (AMH) or antimullerian hormone receptor (AMHR) deficiency;
- testosterone synthesis deficiency due to an enzyme deficiency in the multistep process of testosterone synthesis from cholesterol;
- 5-α-reductase deficiency, whereby no dihydrotesterosterone necessary for action on the androgen receptors is formed from testosterone;
- LH receptor defects, prohibiting normal action of LH.

These are all rare autosomal recessive defects, except the androgen resistance syndrome which is X-linked and more common, and have a frequency of 1:60 000 (Gottlieb *et al*., 1999). These conditions lead to a variable phenotype (from male to ambiguous and even female phenotype) with gonadal dysgenesis and thus azoospermia in most cases. Depending on the phenotype, these individuals will be raised as females or males; if they are raised as males, donor insemination can help them to have children.

(ii) Monogenic defects causing reversible hypogonadism and azoospermia: Luteinizing hormone deficiency, idiopathic hypogonadotropic hypogonadism and Kallmann syndrome all cause hypogonadism and azoospermia. All of these defects can be reversed by adequate hormonal treatment (Buchter *et al*., 1998). Except for the X-linked Kallman syndrome which has a frequency of 1:10 000, these defects are rare (Rugarli, 1999).
(iii) Monogenic defects leading to abnormal sperm parameters in otherwise phenotypically normal males: Primary ciliary dyskinesia and congenital bilateral absence of the vas deferens (CBAVD) due to mutations in the CFTR gene are both autosomal recessive and present respectively with asthenozoospermia (immotile sperm) or obstructive azoospermia (Lissens *et al*., 1996; Cuppens *et al*., 1998).
(iv) Monogenic diseases leading to abnormal sperm parameters and associated problems which allow normal relationship and parenting: For example, Kennedy disease or spinal bulbar muscular atrophy is an X-linked disease that occurs with a frequency of 1:50 000, and results from an androgen receptor defect due to the presence of a CAG trinucleotide repeat expansion (Igarashi *et al*., 1992). Myotonic dystrophy is an autosomal dominant muscular disease that occurs with a frequency of 1:8000 as a result of a CTG expansion in the 3′ region of the DMPK-gene (Mahadevan *et al*., 1992). In addition, as a result of improved therapy, cystic fibrosis—which is an autosomal recessive lung and pancreatic disease occurring with a frequency of 1:2500—is no longer lethal at a young age and young adults do now seek help to procreate. In most of these cases, if spermatozoa are available, ICSI + preimplantation genetic diagnosis (PGD) can be offered to the couples when indicated.
(v) Monogenic diseases leading to sub- or infertility because of severe associated problems, including mental retardation: These include Laurence–Moon–Biedl syndrome, Prader–Willi syndrome, Aarskog syndrome, Beckwith–Wiedemann syndrome, Noonan syndrome, adrenomyeloneuropathy, and others (Liebaers *et al*., 2001).
(vi) Mitochrondrial diseases due to nuclear or mitochondrial gene defects may also play a role in infertility by affecting motility (Liebaers *et al*., 2001).

Male infertility caused by a genetic defect will always be congenital in the sense that the defect is present since conception as an inherited or de-novo anomaly at the chromosomal or DNA level. Although much is known about sex differentiation and spermatogenesis, many questions remain. In the meantime, it is important to be aware in the clinic of the known mechanisms, especially if causal treatment can reverse the situation. Attention

Table III. Risk calculations for a cystic fibrosis (CF) or congenital bilateral absence of the vas deferens (CBAVD) child in case of CBAVD. Amended from Liebaers *et al*. (Liebaers *et al*., 2001).

Investigation	Male	Female	Transmission	Risk
No testing	8/10	1/25	1/4	1/125
Testing female				
Carrier	8/10	1	1/4	1/5
Not a carrier	8/10	1/150	1/4	1/750
Testing male and female				
Female carrier	CF/CF	1	1/2	1/2
Female not a carrier	CF/CF	1/150	1/2	1/300
Female carrier	CF/5T	1	1/4	1/4 (CF) 1/8 (CBAVD)

If the CBAVD patient is not tested for CF mutations, his risk of having at least one CF mutation is 8/10; if his partner is not tested and Caucasian, her risk of being a carrier of one CF mutation is 1/25. A carrier has a risk of 1/2 to transmit the mutation. Two carriers have a risk of 1/4 to transmit their mutated gene at the same time. A CBAVD patient with two mutations will always transmit a mutated gene. Risks for CF can be calculated is none of the partners are tested, if only the female partner is tested, if both partners are tested. In high-risk situations, preimplantation genetic diagnosis (PGD) can be offered.

has also to be paid to those conditions which can be treated by ICSI but may lead to major problems during pregnancy (miscarriages) or at birth (chromosomal or single gene disorders) (Table III). Today, these problems can often be prevented by embryo selection after PGD (Liebaers *et al*., 2001).

Genetic determinants of female infertility

Female infertility is not a well-defined entity. Defined as the lack of conception after an arbitrary period of 12 months, it inadvertently includes a small proportion of normal fertile females who failed to conceive by chance during the 12 or 13 opportunities a woman has per year. Also, the existence of infertility in the female partner does not exclude male causes of subfertility. Of course, fertility can only be expressed by the female partner achieving a pregnancy, and since fertile females are considered to be able to compensate for minor male defects, numerous purely female causes of subfertility remain difficult to study. There exist both non-genetic and genetic causes of female subfertility; the latter are the subject of the present review, though for the sake of brevity, genetic causes of early pregnancy failure and recurrent miscarriage, will not be discussed.

Disorders of the gonadotroph

Disorders in this category are characterized predominantly by hypogonadotrophic hypogonadism (Seminara *et al*., 2000). Apart from low FSH and LH serum levels, women with hypogonadotrophic hypogonadism often exhibit primary amenorrhoea and irreversible pubertal delay. Kallmann's syndrome belongs to this category of disorders. X-linked and autosomal recessive modes of inheritance of hypogonadotrophic hypogonadism have been suggested and have been linked to genetic mutations in up to 10% of patients with low FSH and low LH serum levels. In the majority of cases however, a specific genetic origin remains to be elucidated.

Disorders of female sexual differentiation

Disorders in the development of the gonads may give rise to secondary developmental failures of the genital tract (Ostrer, 2000). Many are characterized by the occurrence of gonadal dysgenesis (Simpson, 1985). Hereditary factors are easily recognized in patients with numerical chromosomal anomalies, such as 45,X or Turner syndrome, but may also be present in patients with 'normal' karyotypes, for example in 46,XY females with the Swyer syndrome (Conway, 2000), with a supposed defect in the testis determining factor (TDF) gene. Androgen insensitivity syndromes (previously called testicular feminization), and androgen synthesis disorders (e.g. 5-α-reductase deficiency and congenital adrenal hyperplasia) also belong to the group of syndromes for which genetic causes are being studied.

Disorders of the uterus and female reproductive tract

A familial preponderance of disorders of Müllerian differentiation (Mayer–Rokitansky–Kuester–Hauser syndrome, incomplete Müllerian fusion syndromes) suggests an origin in polygenic and/or multifactorial inheritance, as does their co-occurrence with, for example, derangements of carbohydrate metabolism, though the location of the affected genes remains to be established. Familial tendencies have been reported for the occurrence of leiomyomas and adenomyosis (Ligon and Morton, 2001). These associations also suggest a polygenic and/or multifactorial inheritance.

Disorders of the ovary

Secondary amenorrhoea with elevated gonadotrophin levels observed under the age of 40 years is defined as premature ovarian failure (POF); however, if it occurs between 41 and 44 years, it is defined as early menopause (EM). The two forms affect 1–2% and 5% of women of the general population respectively. POF is a highly heterogeneous condition and can be associated with autoimmune disorders (see below), ovarian surgery, iatrogenic causes such as chemo-radiotherapy, or with

systemic diseases such as galactosaemia, though in most cases the aetiology of POF is still unknown. A consistent proportion of these patients has a familial form of premature menopause, and pedigree analysis strongly suggests the existence of POF with a dominant maternal and paternal transmission (Vegetti *et al*., 1998). X-chromosomal abnormalities as well as autosomal (recessive and dominant) genes may affect normal ovarian differentiation, maintenance and (accelerated) atresia (as in 45,X patients, and patients with 45,X/46,XX mosaicism). Although the precise location of the affected genes is still largely unknown, it has long been known that proximal regions on the short and long arms of the X-chromosome contain domains that determine ovarian maintenance where deletions often lead to primary amenorrhoea, whereas terminal deletions on the short or long arms of the X-chromosome are more likely to cause POF. Autosomal genes also are involved in ovarian development (for example, the FSH receptor gene), but knowledge of this link is limited.

Fragile X syndrome (FRAXA) is an X-linked dominant condition with incomplete penetrance (Fu *et al*., 1991). The molecular basis of the disease is the expansion of a CTG expansion in the 5′ region of the DMPK-gene (Verkerk *et al*., 1990; Warren and Ashley, 1995). Recent studies indicate an association between POF and FRAXA premutation, and suggest that screening of familial POF for FRAXA premutation will be useful to prevent the transmission of Fragile X syndrome (Marozzi *et al*., 2000). Such screening is particularly relevant when a sister of a patient with POF is being considered as an oocyte donor.

A special place among the ovarian disorders is occupied by PCOS. Although familial clustering occurs, the syndrome is so heterogeneous that genetic factors are very difficult to detect (Legro *et al*., 1998; Govind *et al*., 1999; Legro, 2000). Low penetrance and varying expression of a single gene, as well as polygenic and/or multifactorial inheritance and genetic heterogeneity, have been deduced as determining genetic factors from the findings of innumerable studies. An overabundance of candidate genes has been subjected to linkage analysis, but nearly all have failed to show linkage or association with PCOS. The research in this field has been hampered by the considerable clinical heterogeneity of the syndrome and the lack of a specific pathophysiological defect. Apart from polycystic ovaries, a variety of conditions including hirsutism, anovulation, obesity, insulin resistance, altered gonadotrophin secretion, errors in steroid hormone biosynthesis, hyperandrogenism, hyperinsulinaemia, virilization and impaired fertility have been described as essential elements of PCOS. It becomes increasingly clear however that, in PCOS, the polycystic ovary is a secondary functional derangement and not a specific primary local defect.

Disorders of the pelvic cavity

Family studies have indicated a 2- to 7-fold increase of endometriosis in first-degree relatives (mothers, sisters) of endometriosis patients (Bischoff and Simpson, 2000), with a higher concordance occurring in monozygotic than in dizygotic twins. Localization of genes that might be important in the development of endometriosis has been impeded by two unresolved issues in the development and the clinical picture of the disease. First, it is unclear whether reflux regurgitation of viable menstrual debris by itself leads to the establishment of endometriosis in the pelvic cavity, or whether possibly hereditary immunological factors and inherent deficiencies in the peritoneal lining are responsible for first the attachment and subsequently the invasion and proliferation of endometrial tissue in the parietal and visceral mesothelium. Second, not all investigators agree as to whether endometriosis is the same singular entity in all patients; at least three distinct forms of endometriosis have been discerned by some observers (peritoneal, ovarian and rectovaginal).

The fact that endometriosis remains a superficial quasi-benign disorder in some patients, whereas in others it turns into an aggressive, invasive disease, might help to distinguish factors that endometriosis may have in common with malignant processes. In several studies, familial cases of endometriosis were more likely to show severe endometriosis than non-familial cases. Reporting and detection bias are common, however, and important sources of confounding exist in many of the epidemiological studies reporting on familial clustering of this disease. Clinically, and perhaps also genetically, endometriosis remains a heterogeneous disease. Although the findings from some family studies are highly suggestive of a well-defined hereditary component in some forms of endometriosis, other forms may be polygenic and/or multifactorial, or simply reflect genetic heterogeneity (Campbell and Thomas, 2001). In some groups of patients non-genetic factors might be operative, such as menstrual outflow tract impairment or other disorders of the uterus and the higher female reproductive tract.

The notion that familial clustering exists in some diseases has long since engendered epidemiological observations of hereditary patterns that in turn suggest a sex-linked or autosomal inheritance of an ever-increasing number of gynaecological disorders (Table IV) (Simpson, 2000; Horne *et al*., 2001).

Sexually transmitted diseases

Although the exact incidence of the various factors causing infertility varies among different populations (Gray, 1990) it is generally accepted that 20–30% of cases can be attributed to pelvic factors, such as adhesions from endometriosis and infection, or tubal occlusion which may interfere with normal oocyte retrieval, transport and fertilization (Templeton *et al*., 1991).

The term pelvic inflammatory disease (PID) refers to an infection of the upper genital tract not associated with pregnancy or intraperitoneal pelvic operations. Infertility, ectopic pregnan-

Table IV. Occurrence (%) of disease in first-degree relatives. Amended from Simpson (Simpson, 2000).

Disease	First-degree relatives of:	
	Probands	Controls
Endometriosis	5.5	0.8
Fibroids	21.2	8.4
PCO	30.2	4.2
POF	37.5	9.0

PCO = polycystic ovary syndrome; POF = premature ovarian failure.

cies and chronic pelvic pain, are important consequences of PID, and since sexually transmitted microorganisms are the cause of acute PID in the majority of cases, then PID represents the link between sexually transmitted diseases (STDs) and infertility.

WHO estimates that the number of new cases of STDs among adults in 1999 was approximately 340 million cases. This included gonorrhoea, chlamydial infection, syphilis and trichomoniasis. South and South-East Asia contributed with some 151 million cases, sub-Saharan Africa with 69 million cases, and Latin America and the Caribbean with 38 million cases. In these areas one would expect that considerably more than 30% of infertility cases are attributable to 'pelvic factors' (World Health Organization, 2001).

In more than 99% of cases, acute PID results from bacterial flora that originates in the vagina and cervix and then ascends along the mucosal surface of the uterine cavity and the Fallopian tube, causing endometritis and salpingitis respectively. The infection sometimes extends to the surface of the ovaries and the peritoneum, causing oophoritis, parametritis and pelvic peritonitis. Most of the long-term sequelae of PID however stem from destruction of normal tubal architecture, with or without tubal occlusion (Hager *et al*., 1983).

Although there has been a decline in the incidence of PID over the past few years, at least in western countries, the incidence and cost remain high. There were approximately one million cases in the United States in 1998, with direct and indirect costs estimated as approximately $1.88 billion (Rein *et al*., 2000). Every year, 1–2% of all young, sexually active women suffer from PID, which is the most common serious infection of women aged 16 to 25 years. Approximately 85% of cases appear spontaneously in sexually active females; the remaining 15% follow procedures which interrupt the cervical barrier, such as endometrial biopsy, curettage, intrauterine device insertion, hysterosalpingography, hysteroscopy and voluntary pregnancy termination.

The incidence of infertility following acute PID may vary widely—from 6 to 60%—depending on a variety of factors, such as the number of episodes and the age of the patient. In one study conducted among hospitalized women (Westrom *et al*., 1992), the incidence of tubal obstruction was 11.4% after a single episode, 23.1% after two episodes, and 54.3% after three or more episodes. On the other hand, many women with tubal infertility arising from PID do not have a history of an acute pelvic infection. Over the past decade, this atypical or silent PID—which is defined as an asymptomatic or relatively asymptomatic inflammation of the upper genital tract, and is often associated with chlamydial infection—has become well recognized and is increasingly the subject of investigation (Paavonen, 1998).

Some researchers believe that atypical PID may be the more common form of upper genital tract infection. In women with cervicitis without clinical signs of upper localization, up to 40% have endometritis on endometrial biopsy (Paavonen *et al*., 1985). Likewise, women with acute chlamydial PID and those with silent chlamydial infection have equally severe tubal damage, adhesion, degeneration of endosalpingeal structures and cilia dysfunction (Wolner-Hanssen, 1995). Although a variety of microorganisms have been involved in the aetiology of PID, two classic sexually transmitted organisms, *Neisseria gonorrheae* and *Chlamydia trachomatis*, cause the majority of acute PID cases and coexist in 25–50% of the cases.

C. trachomatis has recently become the most prevalent sexually transmitted microorganism world-wide, with 20–40% of sexually active women having antibodies against it. Between 10 and 30% of women with acute PID have evidence of acute *C. trachomatis* infection by serial antibody testing, although specific cultures remain negative. Finally, approximately 30% of women with acute cervicitis caused by *C. trachomatis* will subsequently develop PID. Endogenous aerobic and anaerobic bacteria originating from the normal vaginal flora are also isolated from tubal fluid in approximately 50% of cases. The polymicrobial flora that is very common throughout the active infectious process, however, may be the consequence of reduced local defences due to colonization by an initial single pathogenic organism.

The type and number of species cultured vary depending on several factors, including the geographical location of the study, the population included, the prevalence of lower genital tract disease, the duration of symptoms, the severity of the disease and the frequency of previous episodes of PID. As to the correlation between endocervical and tubal cultures, according to several laparoscopic studies, the same results are found in no more than 50% of cases (Eschenbach *et al*., 1997).

The woman who is classically at highest risk for STD is a teenager with multiple sexual partners who lives in an area with a high prevalence of STDs and does not use a barrier method of contraception. Among both proven and unproven methods of preventing STDs, PID and consequent infertility, monogamy, reducing the number of sexual partners and the use of barrier and hormonal contraceptive methods seem to be of major importance (Stone *et al*., 1986). As to the role of the intrauterine device (IUD), following early alarmistic studies recent data indicate that the increase in risk for PID occurs only in the first few weeks following insertion (Wolner-Hanssen *et al*., 1985). For this reason, special attention should be paid whenever a transcervical penetration of the mucus barrier is performed for any medical reason. It is noteworthy that approximately 1 out of 200 cases of first-trimester abortion is complicated by an upper genital tract infection (Stenchever *et al*., 2001).

Uterine abnormalities

Normal anatomy of the endometrial cavity is essential for the physiological function of the endometrium. The presence of uterine pathology may cause endometrial dysfunction, impaired implantation and poor pregnancy outcome.

Uterine malformations

Uterine malformations are a group of miscellaneous congenital anomalies of the female genital tract occurring in approximately 4.5% of the general population. Septate uterus is the commonest uterine anomaly (35%), followed by bicornuate uterus (25%) (Grimbizis *et al*., 2001). Although not an infertility factor in itself, the presence of a malformed uterus—especially a septate uterus—is associated with a low term delivery rate (~50%) (Grimbizis *et al*., 2001). In some cases of septate uterus, the pregnancy outcome is even poorer, even from the first pregnancy (Homer *et al*., 2000; Grimbizis *et al*., 2001). Hysteroscopic septum resection is associated with an almost normal prognosis, with term delivery and live birth rates approaching 75% and 85% respectively

(Goldenberg *et al*., 1995; Donnez and Nisole, 1997; Grimbizis *et al*., 1998b; Homer *et al*., 2000; Grimbizis *et al*., 2001).

Submucous myomas

Submucosal uterine myomas are a common uterine pathology which may interfere with embryo implantation, increase abortion and preterm delivery rates or cause complications during pregnancy, such as abnormal placentation and placental abruption (Hamou, 1998; Vercellini *et al*., 1998, 1999; Emanuel *et al*., 1999). Furthermore, submucosal myomas are associated with abnormal uterine bleeding (Emanuel *et al*., 1999; Vercellini *et al*., 1999). While case series remain the highest level of evidence, hysteroscopic myomectomy may be a useful treatment when submucosal myomas are <4 cm in diameter with more than 50% of their volume in the endometrial cavity (type 0 and I) (Vercellini *et al*., 1999). Recurrence of myomas is observed in 7–25% of the patients, occurring more commonly with incomplete resection, type II myomas (5-fold risk) and during the first 2 years after the operation (Emanuel *et al*., 1999; Vercellini *et al*., 1999). The pregnancy rate after hysteroscopic myomectomy in infertile patients is approximately 55% (Bajeral and Li, 2000).

Endometrial adhesions

Adhesion is another serious problem which may be associated with infertility, pregnancy complications such as placenta accreta, and poor pregnancy outcome (Hamou, 1998). The reported incidence in infertile patients is variable, and ranges up to 20% (March, 1995). Adhesions that have been established for less than 3 months are not yet organized and can be easily treated with the distal end of the diagnostic hysteroscope. On the other hand, fibrous well-organized endometrial adhesions can be treated successfully with scissors or the operative resectoscope (Hamou, 1998). Their treatment is associated with pregnancy rates up to 80%, and with a low incidence of pregnancy complications (Stephenson, 1996; Hamou, 1998).

In conclusion, uterine pathology such as septate uterus, submucous myomas and endometrial adhesions may interfere with fertility and pregnancy outcome. Hysteroscopic treatment of these lesions may restore normal uterine anatomy as well as the fertility potential of the patients.

Autoimmunity and human infertility

Females are more susceptible to autoimmune diseases than males, possibly because of oestrogen effects on the normal immune system. Autoimmunity may be associated with premature ovarian failure and recurrent pregnancy loss; an association with infertility remains uncertain.

Premature ovarian failure (see above)

Many cases of premature ovarian failure are unexplained, and it is hypothesized that some of these may arise from ovarian autoimmunity. Although autoantibodies to thyroid peroxidase, thyroglobulin, pancreatic beta cells, adrenal tissues and ovarian tissues may be positive in some cases, only when premature ovarian failure is associated with autoimmune adrenal insufficiency (Addison's disease) is there strong evidence for an autoimmune basis.

Autoimmune adrenal insufficiency often involves other glands, and is referred to as autoimmune polyendocrine syndrome (APS). Type 1 APS is most common in children, and is characterized by Addison's disease, candidiasis and hypoparathyroidism. Ovarian failure occurs in 60% of Type 1 APS cases. Type 2 APS is most common in women aged 30–40 years, and is associated with Addison's disease and hypothyroidism. Ovarian failure occurs in 10% of Type 2 APS cases. Autoimmunity as a mechanism for the premature ovarian failure in APS seems likely because there are autoantibodies to steroid-producing cells and a lymphatic and plasma cell oophoritis (Hoek *et al*., 1997).

Whether premature ovarian failure in the absence of Addison's disease is an autoimmune disorder is less certain (Hoek *et al*., 1997). Insulin-dependent diabetes mellitus and myasthenia gravis are more common with premature ovarian failure, but thyroid and gastric parietal autoantibodies are only slightly more prevalent compared with normal controls. Also, steroid-cell antibodies were not present in premature ovarian failure in the absence of Addison's disease. Moreover, several ovarian autoantibodies were no more prevalent in premature ovarian failure than in normal post-menopausal controls. Furthermore, autoantibodies to ovarian elements such as 3α-hydroxysteroid dehydrogenase vary in prevalence among studies (Reimand *et al*., 2000). Thus, evidence is lacking to establish an autoimmune basis for premature ovarian failure in the absence of Addison's disease.

On rare occasions, women with premature ovarian failure who have a normal karyotype ovulate spontaneously and conceive. On the basis of no better evidence than uncontrolled studies, it is not possible to judge whether glucocorticosteroid treatment is effective for autoimmune ovarian failure.

Recurrent pregnancy loss

Recurrent pregnancy loss (habitual abortion) occurs in approximately 1% of reproductive age couples. Although unexplained in up to half of these of couples, support for an autoimmune mechanism is limited to the presence of antiphospholipid antibodies (APA) in some cases, which is insufficient evidence for a causal association for several reasons. First, the assays for antiphospholipid antibodies other than anticardiolipin are not standardized. Second, studies screen for different APA. Although APA are the most commonly detected autoantibodies in women with recurrent pregnancy loss, antithyroid antibodies, antinuclear antibodies and antiovarian antibodies may be found. In a meta-analysis involving seven studies, APA positivity was based on anticardiolipin or antiphosphatidyl serine presence in one study, three included additional antiphospholipids, and a further three studies included various other autoantibodies (Hornstein *et al*., 2000). Third, there is variability within persons and between assays. Among 500 women with a history of three or more miscarriages, lupus anticoagulant was present in 10% and anticardiolipin antibodies in 3%, but with repeat testing after 8 weeks, only 66% of lupus anticoagulant and 36% of anticardiolipin tests remained positive (Rai *et al*., 2001). A fourth issue is the failure to show consistently that thyroid antibody status affects the future risk of pregnancy loss in women with unexplained recurrent miscarriage (Rushworth *et al*., 2000). Finally, immunotherapy has not been demonstrated to be an effective treatment, while thromboprophylaxis with low-dose aspirin and low-dose heparin is effective in treating recurrent loss

associated with APA, lupus anticoagulant, or anticardiolipin antibodies (Rai *et al*., 1997).

Even in the absence of autoimmune antibodies, an autoimmune basis of unexplained recurrent pregnancy loss has been proposed, hypothesizing that some cases of unexplained recurrent pregnancy loss may arise from an autoimmune barrier to normal placentation. In four randomized controlled trials, however, passive immunization with intravenous immunoglobulin (IVIG) did not significantly improve live birth rates among women with primary unexplained habitual abortion (The German RSA/IVIG Group, 1994; Coulam *et al*., 1995; Perino *et al*., 1997; Stephenson *et al*., 1998).

Infertility

Autoimmunity could be a cause of 'subclinical' ovarian failure in apparently normal women who have elevated basal serum FSH concentrations and poor outcomes after IVF. However, autoantibodies (against ovary, endometrium, thyroid, histones and cardiolipin) and indicators of immune function (CD23, intercellular adhesion molecule concentrations and circulating T-cell subsets) in infertile patients with elevated FSH were not significantly different from infertile patients with normal FSH, nor from laboratory controls. Complement activation was significantly higher among infertile women with elevated FSH and normal FSH, compared with normal controls. The authors concluded that autoimmunity was an infrequent cause of 'subclinical' ovarian failure, but activation of complement might be a characteristic of infertile women (Wheatcroft *et al*., 1997).

The presence of autoimmune antibodies also has been invoked as a possible mechanism of IVF failure. The supposed mechanisms would include abnormal implantation or placentation, and early embryonic vascular compromise. In a meta-analysis of seven studies involving 2053 patients however, antiphospholipid antibody abnormalities did not affect the IVF clinical pregnancy rate (OR 1.0, 95% CI 0.6–1.5) (Hornstein *et al*., 2000). A later report confirmed the lack of association (Chilcott *et al*., 2000). The authors of the meta-analysis concluded that the measurement of antiphospholipid antibody concentrations was not indicated in patients undergoing IVF. Based on a similar autoimmune hypothesis, IVIG and antithrombogenic therapy with aspirin and heparin have been proposed as empiric treatments for IVF failure. No statistically significant differences were detected in implantation, pregnancy and ongoing pregnancy rates between those who received standard therapy and those treated with heparin and aspirin (Kutteh *et al*., 1997).

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