In Vitro Fertilisation: Perinatal Risks and Early Childhood Outcomes

1. Introduction

It is estimated that infertility affects one in seven couples in the UK at some stage in their reproductive life. After over 30 years of experience, in vitro fertilisation (IVF) is now routine medical practice in the management of infertility, and births following IVF are estimated to account for over 1% of all births in the UK. This proportion is notably lower than some European countries, reflecting the different policies of public funding for this procedure.

This paper summarises the growing body of evidence concerning the outcomes of pregnancy, delivery and early childhood associated with being conceived by IVF and related procedures. For these purposes this review will include data relating to IVF and associated procedures such as intracytoplasmic sperm injection (ICSI), blastocyst culture, assisted hatching and genetic diagnosis but will not cover gamete intrafallopian transfer (GIFT) or ovulation induction alone or with artificial insemination. Unless specifically stated, the term IVF is used to encompass IVF and any laboratory procedure associated with IVF.

2. Perinatal risks

2.1 Multiple conceptions and multiple births

The single most important determinant of pregnancy and long-term outcome is whether the pregnancy is a singleton or multiple gestation, irrespective of whether it is a natural or assisted conception. Furthermore, the risks increase disproportionately in relation to the number of fetuses, with monozygosity also being associated with higher rates of adverse outcomes. IVF itself appears to increase the risk of monozygous twins by two-fold compared with natural conception, although the incidence of monozygotic twins even after IVF remains very low. At present, about one in four of all IVF pregnancies result in a multiple birth in the UK owing to the common practice of replacing two or three embryos, with the vast majority of multiple pregnancies occurring after multiple embryo transfer. Multiple pregnancy is perceived as an ideal outcome by many parents and indeed for the majority of multiples, particularly twins, there are no long-term adverse consequences. However, at present it is impossible to predict who will or will not have complications perinatally or later in life. Consequently, elective single embryo transfer and limitation in the UK to the transfer of two embryos in women under the age of 40 is a population level strategy to minimise the risks of prematurity and its associated complications and socioeconomic costs. The aim of this strategy is to reduce IVF-related multiple pregnancy to less than 10% by 2012. When multiple pregnancy is diagnosed in a fertility unit, referral to a specialised multiple birth clinic should be made. This would allow the couple to receive counselling about selective fetal reduction for triplets and high order pregnancies, as well as specialist prenatal screening and diagnosis.

2.2 Preterm birth

Multiple pregnancy per se is a clear risk factor for preterm birth; however, there is an additional small but statistically significant 23% increase in the relative risk of preterm birth in IVF twins compared with natural twins, although the relative contribution of spontaneous or elective preterm birth has not been identified. Similarly, although in singletons there is an estimated two-fold increased risk of preterm birth and moderate preterm birth following IVF, the contribution of spontaneous preterm labour is unknown. Clearly, spontaneous and elective preterm births have largely different aetiologies, principally reflecting infection and placental dysfunction, respectively. Consequently, maternal and treatment factors which are associated with these will have an impact on the overall outcome. For example, duration and cause of infertility can influence the risk of preterm birth and treatment characteristics. Treatment requiring donor oocytes or ICSI can influence the risk. In addition, early fetal loss in a multiple gestation can increase the risk of preterm birth for the remaining singleton. Although
many of these factors cannot be addressed, accurate estimates of risk will improve counselling and increase the likelihood of the mother or couple choosing elective single embryo transfer.

2.3 Low birth weight and small for gestational age

Although prematurity due to multiple pregnancy is a clear risk factor for low birth weight (LBW) <2500 g, very low birth weight (VLBW) <1500 g and extremely low birth weight (ELBW) <1000 g, singleton IVF pregnancies still demonstrate an increased risk of low birth weight compared with naturally conceived singletons (RR 1.6 95% CI 1.29–1.98; RR 2.65, 95% CI 1.83–3.84; RR 3.02, 95% CI 0.12–74.66 for each birth weight category respectively). Furthermore, in IVF twin pregnancies, this increased risk is still evident compared with natural twins (LBW RR 1.14, 95% CI 1.06–1.22, VLBW RR 1.28, 95% CI 0.73–2.24 and ELBW RR 0.88, 95% CI 0.04–19.40). Although prematurity will partly influence the risk of LBW in singletons, the relative risk of babies being small for gestational age is also increased by approximately 40–60%, suggesting that factors other than preterm birth are responsible. Notably, the number of original fetal hearts observed on ultrasound is a determinant of the risk of LBW, and inevitably the risk of impaired fetal growth will be partially ameliorated by elective single embryo transfer. However, the characteristics of the mother and father and the treatment cycle may also influence the risk of LBW and these will be harder to address.

2.4 Congenital anomalies

Between 3% and 5% of all infants are diagnosed with a congenital anomaly soon after birth. IVF is associated with a 30–40% increased risk of major congenital anomalies compared with natural conceptions. Notably, this risk is not attributable to the increased risk of congenital anomalies associated with multiple birth, as even in singleton pregnancies the excess risk remains. It appears that the increased risk is partly attributable to the underlying infertility or its determinants as couples who take longer than 12 months to conceive also exhibit an increased risk of anomalies (hazard ratio [HR] 1.20, 95% CI 1.07–1.35), although this was not as high as that observed in treated infertile couples (HR 1.39, 95% CI 1.23–1.57). The principal anomalies which occur in IVF pregnancies include a range of gastrointestinal, cardiovascular and musculoskeletal defects and specifically septal heart defects, cleft lip, oesophageal atresia and anorectal atresia. Of note, while the relative risk of major congenital anomalies associated with IVF is in the order of 30–40%, the absolute risk is nevertheless low since anomalies per se are relatively uncommon.

2.5 Vertical transmission of genetic diseases

In some cases infertility may be genetic in origin, and successful IVF treatment may therefore facilitate intergenerational transmission. This has led to concern that children born following these techniques will express a greater number of genetic abnormalities. There is an increased prevalence of structural chromosomal abnormalities in infertile men and women: a 4.6% prevalence of autosomal translocation and inversions in oligospermic men, and a 1.14% prevalence of autosomal reciprocal balanced translocations in infertile women (general population 0.16%). However, these structural abnormalities would normally be detected and subsequent transmission avoided by use of preimplantation genetic diagnosis (PGD) following karyotyping of men with azoospermia or severe oligospermia, and in couples with recurrent implantation failure or miscarriage. Microdeletions of the long arm of the Y chromosome (Yq), in particular the AZF region, can also cause spermatogenic failure and either oligo- or azoospermia, the latter preventing further vertical transmission. However, sons conceived from oligospermic men with Yq microdeletions will inherit this subfertile phenotype, and further expansion or de novo deletions may occur resulting in a worse phenotype in the offspring. Although other single gene disorders such as cystic fibrosis are associated with infertility, owing to congenital absence of the vas deferens, vertical transmission of the common mutations can be avoided through testing the female partner and performing PGD if she is a carrier. Lastly, there is increasing evidence that epigenetics may contribute to abnormal embryo and trophoblast development, with IVF
superovulation and culture conditions capable of inducing epigenetic changes and long-term genomic imprinting. To date, nine human imprinting syndromes have been identified, but current evidence links IVF with only three: Beckwith-Wiedemann syndrome (BWS), Angelman syndrome (AS) and, more recently, maternal hypomethylation syndrome. The overall incidence of these conditions is very low at less than 1 in 12,000 births (BWS 1 in 13,700, and Angelman syndrome 1 in 16,000) and consequently routine screening for imprinting disorders in children born after IVF is not recommended. Although these conditions may represent the extreme end of the epigenetic spectrum, there are potentially much subtler effects as the type of culture media used can profoundly affect birth weight in humans and animals – the so-called large offspring syndrome in cattle and sheep. Similarly, administration of exogenous hormones to induce oocyte maturation in vivo, as well as in vitro, may hamper the proper acquisition of maternal imprints during oogenesis and both maternal and paternal methylation-dependent imprints during embryo development in a dose-dependent manner.

2.6 Perinatal mortality

Some caution should be taken in the interpretation of the pooled findings relating to perinatal mortality and IVF because of a particular lack of consistency in results across the studies pooled and the influence of one particularly large study. However, accepting this, the overall pooled result suggests there is nearly a 70% increase in the risk of perinatal death for IVF singletons compared with natural conceptions. A recent prospective follow-up study of 20,166 singleton pregnancies compared the risk of stillbirths between fertile women, subfertile women (time to natural conception greater than 12 months) and women pregnant after fertility treatment (IVF and non-IVF assisted reproductive technique [ART]). Only women who conceived with IVF had a statistically significant four-fold increased risk of stillbirth compared with fertile women. This would suggest that the increased risk of stillbirth is associated with treatment-related factors to a greater degree than infertility/subfertility itself. However, this is in contrast to the findings from three studies of perinatal mortality in singletons conceived naturally following a prolonged time to conception, which found a two- to three-fold increased risk of perinatal death compared with singletons naturally conceived without a prolonged time to conception. The results for twins are similarly inconsistent, but the combined results do suggest that, having adjusted for confounders, there is a decrease in the risk of perinatal death in IVF twins of between 16% and 42% compared with natural twins, potentially reflecting the lower incidence of monozygotic twins after IVF.

3. Specific IVF and related procedures

3.1 Preimplantation genetic diagnosis/screening

Preimplantation genetic diagnosis/screening (PGD/PGS) requires the removal of one or more blastomeres from the embryo for genetic testing, thereby allowing the transfer of unaffected embryos. Despite the removal of a variable amount of material from the developing embryo, the overall prospect for children appears in line with standard ICSI. However, PGD/PGS has been associated with a substantial increased risk of perinatal death compared with standard ICSI (4.6% versus 1.9%; OR 2.56). When stratified for multiple births, perinatal death rates among PGD/PGS singleton and ICSI singleton children were similar (1% versus 1.3%), but there were significantly more perinatal deaths seen in post-PGD/PGS multiple pregnancies compared with conventional ICSI multiple pregnancies (11.7% versus 2.5%), and this was not attributable to an increased prevalence of monozygotic twins. Notably, all of these perinatal mortality rates are substantially higher than the general population rates, and further research is required to elucidate the time of death to determine whether timing of induction of labour could be altered to minimise the risk at term.

3.2 Blastocyst culture

The extended culture of embryos in vitro from the traditional day 2 (cleavage-stage) transfer date to day 5 (blastocyst stage) has been advocated as a means of improving embryo selection and thus pregnancy rates. The embryo is sequentially cultured in different media to facilitate in vitro development. In a
A Cochrane review, the evidence of a significant difference in live-birth rate per couple between the two treatment groups was detected in favour of blastocyst culture (day 2/3: 29.4% versus day 5/6: 36.0%).

A systematic review and meta-analysis has demonstrated that the probability of live birth after fresh IVF is 40% higher after blastocyst-stage embryo transfer compared with cleavage-stage embryo transfer when equal number of embryos are transferred.

Studies in different animal models have raised some concerns about potential deleterious effects of extended embryo culture, including imprinting disorders, overgrowth, behavioural abnormalities as well as cardiovascular and metabolic dysfunction. In the human, a sex ratio in favour of males has been associated with blastocyst culture. After adjusting for confounders, the risk of preterm birth among singletons was significantly greater after blastocyst-stage transfer than after cleavage-stage transfer; the risk of congenital malformations was also significantly higher. The odds of any congenital malformation versus population figures was 50% higher after blastocyst transfer whereas after cleavage-stage transfer it was only 11% higher. These differences in outcome may be attributable to an increase in monozygotic twinning; however, there are conflicting data regarding blastocyst transfer and the risk of monozygotic twinning. A retrospective study has demonstrated that monozygotic twinning is not increased after single blastocyst transfer compared with single cleavage-stage embryo transfer, whereas other authors have concluded that blastocyst culture is associated with an increased risk of monozygotic twinning. These differences might be related to a variation in culture media. Elective single blastocyst transfer is an effective option to decrease the risk of multiple birth while maintaining a high pregnancy rate. Nevertheless, the possible risks associated with the extended period in culture require continuing surveillance of the children born after blastocyst transfer.

### 3.3 Assisted hatching

Assisted hatching (AH), a technique whereby the zona pellucida is disrupted to facilitate implantation, has recently been shown to enhance clinical pregnancy rates (OR 1.29, 95% CI 1.12–1.49) but not live birth (OR 1.13, 95% CI 0.83–1.55) nor to reduce multiple pregnancy rates (OR 1.67, 95% CI 1.24–2.26). The long-term impact on the pregnancy and offspring is, however, unknown.

### 3.4 In vitro maturation

Immature oocyte retrieval and subsequent oocyte maturation in vitro (IVM) without any ovarian stimulation is a new development in ART. IVM gives the benefits of ovarian stimulation – namely, more oocytes – without the risks of ovarian stimulation. Patients with polycystic ovary syndrome (PCOS) or polycystic ovaries represent a good indication for IVM. Over 1000 babies have been born worldwide following IVM and, although formal prospective paediatric follow-up studies are limited, similar obstetric outcomes and congenital anomaly rates between babies born following IVM, IVF and ICSI have been reported. Neurological development of IVM children at 2 years of age also appears to be normal, although this conclusion is based on only 67 children.

### 4. Long-term outcomes for children

The physical, neurological and developmental health of children born after IVF is certainly one of the most important aspects when discussing the potential adverse effects of IVF. Overall, the neuromotor, cognitive, language and behavioural outcomes in children born following IVF or ICSI appears to be similar to children conceived naturally. The only consistent adverse finding has been an increased risk of cerebral palsy, which is partly, but not wholly, explained by the increased risk of preterm delivery (unadjusted OR 2.18, 95% CI 1.71–2.77). With respect to children’s growth, there appears to be little impact at age 12.

The Barker hypothesis predicts that adverse antenatal conditions can lead to long-term consequences in the adult. For example, under-nutrition during pregnancy is associated with an increased risk of coronary heart disease, type 2 diabetes, stroke and hypertension. It is therefore legitimate to question if IVF might
be associated with cardiometabolic disturbances occurring in adulthood or even earlier during adolescence. Ceelen et al. have observed that peripheral adipose tissue mass was higher in IVF children. The same authors have also reported cardiometabolic differences in children born after IVF, with approximately a 4 mmHg and 2 mmHg increase in systolic and diastolic blood pressure respectively and an increase in fasting glucose, with none of these differences explained by confounders including current body size, birth weight, and other early life factors or by parental characteristics, including subfertility cause. Moreover, these authors have suggested that early childhood growth (weight gain) might predict cardiovascular risk factors (blood pressure and body fat composition) in IVF children. These important data strongly emphasise the need for metabolic epidemiological studies in IVF adolescents and adults.

5. Maternal morbidity

5.1 Maternal age

Increasing maternal age is a risk factor for almost all pregnancy and perinatal complications. The average age at which women attempt to conceive continues to rise and consequently IVF is increasingly used by older women who are already predisposed to pregnancy complications. However, even when comparing age-matched controls there appears to be an increased risk of complications associated with infertility, with a higher rate of caesarean section delivery, obstetric haemorrhage, pre-eclampsia, pregnancy-induced hypertension and gestational diabetes all noted in older women having IVF. All of these conditions are associated with unfavourable perinatal outcomes for the neonate, including preterm delivery, low birth weight and admission for neonatal intensive care. By virtue of their age, older women are more likely to have pre-existing comorbidities further complicating their pregnancy course and outcome. Importantly, women with significant comorbidities, regardless of their age, should receive pre-IVF assessment and counselling.

5.2 Recipients of donor oocytes

With delays in the age at which women attempt to conceive, an increasing number of women are using oocyte donation, a technique previously limited to women with premature ovarian failure. Again there appears to be an increase in early pregnancy and perinatal complications, with the risk of pregnancy-induced hypertension in particular ranging from 16% to 40% of cases, with the greatest risk observed in primiparous women. Despite these risks, there is almost no information on the long-term outcomes of egg donation pregnancies for the mother and her child.

5.3 Polycystic ovary syndrome

PCOS, as defined by the 2003 Rotterdam criteria, is a common condition affecting 6–10% of reproductive-aged women. Metabolic syndrome is often encountered in PCOS and obesity is a classic clinical manifestation. Long-term risks include an increased risk of type 2 diabetes and there is some evidence of an increased risk of cardiovascular events. In these women, the presence of infertility caused by anovulation often requires treatment such as ovulation induction with clomifene citrate, ovarian stimulation with gonadotrophin and IVF. Pregnancy outcomes in women with PCOS were analysed in a meta-analysis. The results indicate an increased risk of gestational diabetes (OR 2.94, 95% CI 1.70–5.08), pregnancy-induced hypertension (OR 3.67, 95% CI 1.98–6.81), pre-eclampsia (OR 3.47, 95% CI 1.95–6.17) and preterm birth (OR 1.75, 95% CI 1.16–2.62). In addition, an increased risk of admission to neonatal intensive care (OR 2.31, 95% CI: 1.25–4.26) and higher perinatal mortality (OR 3.07, 95% CI 1.03–9.21), which were not related to multiple births, have been observed. These data highlight the importance of pre-ART counselling of women with PCOS and emphasise the need for weight management before ART.

6. Antenatal screening

Use of first-trimester combined ultrasound and biochemical screening for Down syndrome is now recommended; however, the effect of IVF is largely ignored. Notably, pregnancy-associated plasma
protein-A (PAPP-A) levels are significantly lower in fresh transfer IVF pregnancies, the consequence of which is an increased risk of receiving a false-positive result and increased odds of having a chorionic villous sampling or amniocentesis.\textsuperscript{79,80} However, this increase in false-positive rate was not confirmed in a recent, smaller, study.\textsuperscript{81} This may reflect the relative inaccuracy of ultrasound dating, with the precise dating available for IVF pregnancies being largely ignored.\textsuperscript{82-84} Further larger studies will be required before recommendations for adjustment of risk calculations for IVF pregnancies can be made.

7. Contribution of the subfertile phenotype

Although many of the above studies have used normal controls as the comparator, it is increasingly clear that the factors which predispose to infertility are also linked with adverse perinatal outcomes, with subfertility acting as a proxy for this. Therefore, in the future researchers trying to determine the individual effect of a novel technique or infertility treatment should consider also using women with a prolonged time to natural conception as the comparison group.\textsuperscript{80,81} This will allow delineation of whether the IVF procedure itself or factors inherent to the couple and their infertility are associated with any adverse outcome.

8. Opinion

While it is clear that IVF pregnancies are at increased risk of adverse perinatal outcomes, it is also the case that the majority of the children born following IVF will have a good outcome. For those with poorer outcomes this inevitably reflects aspects of the treatment but also the interplay with the underlying features that the couple bring to the pregnancy. Further research is needed to untangle this complex relationship to allow effective targeted interventions. Given the known risks associated with IVF pregnancies, risk assessment is required during antenatal care with appropriate referral.\textsuperscript{86} The adoption of elective single embryo transfer provides a clear example of how treatment strategies can be altered to improve outcomes. At present the long-term follow-up studies on children born following IVF are largely reassuring once the confounding factors of prematurity and multiple gestation are removed. However, with the continued refinement of the technical process and clinical application of novel developments, continued surveillance is a prerequisite.

References


